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HEREDITY VOLUME 4 PART 3 DECEMBER 1950

SELECTION IN THE POLYMORPHIC LAND SNAIL *CEPÆA NEMORALIS*

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Received 31.i.50

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INTRODUCTION

THE structure of natural populations of animals and plants and of variation both within and between colonies needs more study as Diver (1940) has pointed out. Research along these lines is of particular importance because of the evidence he has obtained that many, apparently continuous, natural populations are broken up into small breeding-communities, giving the conditions in which genetic drift, as suggested by Sewall Wright (*e.g.* 1940), might be expected to occur if it were operative. Diver has stated that from his observations on populations of *Cepæa nemoralis* (L.) and *C. hortensis* (Müll.) he obtained no indications that the variations in shell colour and banding have any selective value. He considers that these small breeding-communities show definite divergences from each other because of non-random mating, but expresses surprise (1940, p. 312) that such divergences have not proceeded further by means of genetical drift.

The purpose of this paper is to describe an investigation into this problem of the relative importance of selection and drift in determining the distribution of different colour and banding patterns in *C. nemoralis*. We find, on the contrary, that they have definite selective values, related to the environment, determining the general aspect of different populations and therefore of their gene ratios. Although small inbreeding communities within each population may diverge from one another in colour and banding patterns, this random divergence cannot proceed far, because of the effects of natural selection.

METHODS

(i) *Collecting*

Collections were taken from various localities in southern England, situated, with one exception, within a fifty-mile radius of Oxford,

chiefly to the south and west. The localities were chosen for diversity of vegetation, and when possible several widely separated colonies on the same type of vegetational background were examined. Fig. 1 is a sketch-map showing the localities. The dryness of the summer (1949) has made the collection of large samples very difficult, as the animals have remained buried for long periods.

FIG. 1.—Sketch map showing the collecting localities. The broken line represents the 500-foot contour. Steep chalk scarps from which collections were taken are shaded.

Localities :

1. Urchfont Hanger
2. Broomsgrove
3. Cobham Frith
4. Putall Gate
5. Rough Down
6. Rockley 1
7. Rockley 4
8. Hackpen
9. Burdrop Wood
10. Wantage 1
11. Wantage 2
12. East Ilsley
13. Streatley South Wood
14. Sowberry Court
15. The Crown, Huntercombe
16. Christmas Common (Wood and Downside)
17. Shirburn Lodge
18. Aston Rowant
19. Bagley Wood
20. Hinksey Hedge
21. Wytham Lane
22. Wytham Woods
23. Wytham Woods
24. Canal Bank
25. Parks Hedge

In each locality, all the shells found in several small areas within the locality were collected, whether alive or dead, whole or broken, except that no shells less than half the adult size were taken (in fact, very few that were not adult) and that fragments were taken only when the complete band-formula could be determined from them, and it was obvious that each piece was derived from a different shell.

(ii) *Recording*

To give as much information on each shell as was practicable, von Martens' system (1832) of band-formulæ was adopted with slight modifications, since more detailed systems were found to require disproportionate labour in recording. The bands are numbered 1 to 5, the uppermost being 1. Thus a five-banded shell is represented by 12345, a shell with the middle band only as 00300, and one with

no bands as ooooo. Fusions are shown by bracketing the fused bands. As fusion may begin at almost any time in the growth of the shell, an arbitrary distinction must be made to avoid undue complexities. All bands fused at and after a line drawn across the body-whorl from the umbilicus at right angles to the lower lip of the mouth were considered as fused. This convention is illustrated in fig. 2. In a few specimens (all of them with brown shells) the bands could not be distinguished from the general shell-colour; these were classed as "banding indeterminable." Weakly pigmented bands which are interrupted in several places were represented by colons, and were considered as bands for the purpose of this paper. Bands present only as traces close to the lip were recorded as traces but disregarded for the classification employed here.

12345 123(45)

FIG. 2.—Convention used in classifying partial fusing. If fusion extended from the lip to the line shown or beyond, the bands were considered fused.

Ground colour was recorded as approximating to one or other of a series of shells chosen as colour standards and representing the commonest colours. The colour of the intact shell is either yellow or belongs to one of two series, both beginning with a fawn-brown, one leading to a deep sepia-brown, the other through orange-brown and dull pink to a dull red. These colours are the result of pigment in the calcareous layers of the shell showing through a thin, slightly brownish, yellow periostracum. Yellow shells have varying amounts of yellow pigment, medium brown and deep brown ones have a dull leaden violet. All the others have pink. We therefore distinguished three main colour groups, with yellow, violet, and pink-pigmented shells.

Some very pale fawn shells approach the yellow group, which otherwise is distinct and has not been subdivided. The pink series was broken up into three classes, fawns, pinks, and reds. Intermediates were placed in the class they most resemble. The browns vary from medium to dark, but subdivision has not been found necessary. Old shells that had lost the periostracum have been recorded separately.

It was noticed that as Taylor (1907-14) has pointed out, the ground colour of the shell is paler when there are several bands present than when there are few.

TABLE 1

Collections classified by ha

Locality	12345	(12)345	1(23)45	123(45)	(123)45	1(2345)	(12345)	(12)3(45)	1(23)(45)	(123)(45)	(12)(345)	02345	023(45)
Downland Beech—													
Hackpen	...	...	...	...	...	...	...	...	...	...	...	...	...
Rockley 1	...	...	...	...	...	...	...	...	...	...	...	...	...
Rockley 4	...	...	...	...	...	...	...	...	...	...	...	...	...
Crown Huntercombe	2	...	...	...	...	...	...	...	...	...	...	...	...
C. C. Wood	5	...	...	...	...	...	...	2	1	...	...	...	...
Aston Rowant	...	...	...	...	...	...	...	...	...	...	...	...	...
Shirburn Lodge	...	...	...	...	...	...	...	...	...	...	...	...	...
Urchfont Hanger	...	...	...	...	...	...	...	...	...	...	...	...	...
Total	7	...	...	...	...	...	...	2	1	...	...	...	...
Oakwood—													
Broomsgrove	6	...	...	...	...	...	6	...	...	...	...	...	...
Cobham Frith	10	...	1	3	1	...	12	1	...	...	...	...	...
Putall Gate	68	...	...	1	...	...	69	64	...	...	...	1	1
Total	84	...	1	4	1	...	87	65	...	...	...	1	1
Mixed Deciduous Wood—													
Wantage 1	4	...	...	...	...	...	1	4	...	...	...	...	...
Streatley S. Wood	20	...	2	...	...	...	...	2	...	...	...	...	...
Wytham	108	6	5	8	...	...	...	15	...	1	1	18	3
Bagley	13	1	...	2	1	...	...	1	...	...	...	1	...
Burdrop Wood	37	...	1	3	...	...	...	...	...	...	...	9	3
Wantage 2	15	2	...	1	...	...	2	9	...	1	...	...	...
Total	197	9	8	14	1	...	3	31	...	2	1	28	6
Hedgerows—													
Wytham Lane	13	...	...	...	...	...	1	1	...	...	...	...	...
Canal Bank	105	4	5	14	1	1	16	16	3	18	...	...	...
Parks Hedge	57	6	1	2	...	...	...	12	...	...	...	...	...
Hinksey Hedge	14	...	...	5	...	...	...	4	...	...	...	2	...
Total	189	10	6	21	1	1	17	33	3	18	...	2	...
Downside, Long Coarse Grass—													
Pentridge	283	5	4	4	...	...	7	6	...	...	...	4	...
C. C. Downside	54	1	...	13	...	...	...	9	...	...	1	11	5
Total	337	6	4	17	...	...	7	15	...	...	1	15	5
Downside, Short Turf—													
Rough Down	...	...	...	...	...	...	8	...	...	...	...	...	...
Other Stations—													
E. Ilsley	9	...	...	...	...	...	...	...	...	...	...	...	...
Sowberry Court	7	1	1	8	...	...	2	4	1	1	...	...	...
Totals	830	26	20	64	3	1	124	150	5	21	2	46	12

by banding

02(345)	0(2345)	0(23)(45)	10345	12045	12340	10045	02340	00345	003(45)	00(345)	00045	000(45)	00340	00300	00000	Indeterminate	Total
...	...	...	...	...	...	...	...	...	...	...	...	...	...	65	94	...	159
...	...	...	...	...	...	...	...	...	...	...	...	...	...	41	115	...	156
...	...	...	...	...	...	...	...	...	...	...	...	...	...	2	3	...	5
...	...	...	...	...	...	...	...	...	...	...	...	...	...	6	3	...	11
...	...	...	...	...	...	...	...	1	...	...	...	...	...	10	7	...	26
...	...	...	...	...	...	...	...	...	...	...	...	...	...	...	11	...	11
...	...	...	...	...	...	...	...	...	...	...	...	...	...	1	6	...	7
...	...	...	...	...	...	...	...	...	...	...	...	...	...	2	4	...	6
...	...	...	...	...	...	...	...	1	...	...	...	...	...	127	243	...	381
...	...	...	...	...	...	...	...	...	...	...	...	...	...	49	36	...	97
...	...	...	...	...	...	...	...	...	...	...	...	...	...	117	54	...	199
...	...	...	2	...	...	...	...	7	...	...	...	...	...	185	211	...	609
...	...	...	2	...	...	...	...	7	...	...	...	...	...	351	301	...	905
...	...	...	...	...	...	...	...	...	...	...	...	...	...	...	18	...	27
...	...	...	...	...	...	...	...	...	...	...	...	...	...	5	44	...	73
...	...	...	5	1	1	...	3	16	5	...	5	2	4	31	38	2	278
...	...	...	1	...	...	...	...	...	...	...	...	...	...	5	10	...	35
...	1	...	1	...	...	...	1	5	...	...	...	...	...	9	44	...	114
...	...	...	...	...	...	...	...	...	...	...	...	...	...	...	22	...	52
...	1	...	7	1	1	...	4	21	5	...	5	2	4	50	176	2	579
1	...	...	2	...	...	...	...	1	...	...	...	...	...	1	4	...	24
...	...	...	...	...	...	...	...	7	2	...	1	...	...	53	33	...	279
...	...	...	...	...	...	...	...	...	...	...	...	...	...	2	1	...	81
...	...	...	...	...	...	...	...	1	...	...	...	...	...	6	9	...	41
1	...	..	2	...	...	...	...	9	2	...	1	...	...	62	47	...	425
...	...	...	9	...	...	...	...	4	...	...	...	...	...	102	4	2	434
...	1	...	6	...	...	1	...	17	2	...	...	1	...	17	19	...	158
...	1	...	15	...	...	1	...	21	2	...	...	1	...	119	23	2	592
2	...	1	...	...	...	...	...	2	2	2	...	...	1	45	101	...	164
...	...	...	...	...	...	...	...	1	...	...	...	...	1	5	23	4	43
...	...	...	...	...	...	...	...	...	...	...	...	...	...	...	45	...	70
3	2	1	26	1	1	1	4	62	11	2	6	3	6	759	959	8	3159

(The colour divisions used above appear to correspond to the varieties described or illustrated by Taylor as follows :—

Yellows are all var. *libellula* Risso.
Fawns are var. *petiveria* Moquin-Tandon.
Pinks are var. *rubella* Picard in the strict sense.
Reds are var. *rubra* Baudon.
Browns include var. *castanea* Picard and var. *olivacea* Risso except that var. *petiveria* Moquin-Tandon is reduced by Taylor to a subvariety of var. *castanea* Picard in the wider sense. As its shell pigment is pink, not violet, it should be retained as a separate variety.

RESULTS

The results of our collecting are shown in tables 1 and 2. In these, the localities have been classified according to their broad ecological features. We have collected in six main types of habitat, downland beechwoods, oakwoods, mixed deciduous woods, hedgerows, downsides with long coarse herbage, and downsides with very short turf. Two localities not referable to any of these are listed separately.

The downland beechwoods are all pure beech. The woods at Aston Rowant, Shirburn Lodge and Christmas Common are dense, with a thick permanent carpet of tough red-brown beech leaves. Christmas Common and Aston Rowant have a small quantity of brambles (*Rubus* sp.). Undergrowth in dense beechwoods is very slight. The woods at Hackpen and Rockley 1 are more open, with much thin sparse grass, and an incomplete leaf-carpet with exposures of dark brown soil and small stones of chalk and flint. Rockley 1 is notable for the large quantity of fallen beech cupules (which are very dark brown). Hackpen and Rockley are in exposed situations, and wind is responsible for the incompleteness of the leaf-carpet. Urchfont Hanger has large drifts of leaves, alternating with bare areas. The collection from The Crown, Huntercombe, was taken from the edge of a beechwood, on a steep, dry, rather bare roadside bank overshadowed by the trees.

It will be seen from the tables that the average proportion of unbanded and one-banded shells is far higher in all the beechwoods investigated than in any other type of locality. The average proportion of yellow shells is much less than in downland grass or hedgerows, but higher than in oakwoods or in the two unclassified localities. It is rather similar to that for mixed deciduous woods, but it is almost entirely due to the collections from Hackpen and Rockley 1, which were rather open with some grass.

The oakwoods visited differ in density. Broomsgrove is a very dense oakwood with etiolated hazel shrubs and nothing but dead, brown, leaves on the ground. Cobham Frith was like Broomsgrove

TABLE 2

Collection classified by colour

Locality	Fresh shells					Old shells			Total
	Yellow	Fawn	Pink	Red	Brown	Colour unknown	Pink	Yellow	
Downland Beech—									
Hackpen	39	51	49	15	2	3	...	...	159
Rockley 1	42	14	25	9	66	...	...	...	156
Rockley 4	...	...	...	1	4	...	...	...	5
Crown Huntercombe	...	3	3	3	...	...	2	...	11
C.C. Wood	...	...	...	...	...	8	15	3	26
Aston Rowant	...	4	4	3	...	...	...	...	11
Shirburn Lodge	...	2	3	1	...	...	1	...	7
Urchfont Hanger	1	...	3	2	...	...	...	...	6
Total	82	74	87	34	72	11	18	3	381
Oakwood—									
Broomsgrove	7	5	27	...	47	...	8	3	97
Cobham Frith	25	24	118	12	6	...	13	1	199
Putall Gate	22	293	174	33	59	...	28	...	609
Total	54	322	319	45	112	...	49	4	905
Mixed Deciduous Wood									
Wantage 1	8	15	...	...	3	...	...	1	27
Streatley S. Wood	6	15	33	4	8	...	5	2	73
Wytham	69	84	37	3	28	16	41	...	278
Bagley	6	8	15	2	4	...	...	...	35
Burdrop Wood	37	15	22	1	24	3	5	7	114
Wantage 2	14	1	29	...	8	...	...	...	52
Total	140	138	136	10	75	19	51	10	579
Hedgerows—									
Wytham Lane	16	3	1	...	1	1	2	...	24
Canal Bank	114	101	53	...	11	...	...	...	279
Parks Hedge	14	46	16	...	...	2	3	...	81
Hinksey Hedge	18	7	6	...	...	7	3	...	41
Total	162	157	76	...	12	10	8	...	425
Downside, Long Coarse Grass—									
Pentridge	277	1	127	...	26	...	3	...	434
C. C. Downside	66	45	43	1	...	1	2	...	158
Total	343	46	170	1	26	1	5	...	592
Downside, Short Turf—									
Rough Down	74	8	17	...	65	...	...	...	164
Other Localities—									
E. Ilsley	6	1	...	...	35	1	...	...	43
Sowberry Court	7	31	...	...	32	...	...	...	70
Totals	868	777	805	90	429	42	131	17	3159

but a large part was felled about four years ago, with a consequent invasion of rosebay willow-herb (*Epilobium angustifolium* L.) and some brambles. The collection was taken from this part. The willow-herb forms dense thickets of straight and very leafy stems about five feet high in summer. Putall Gate is rather more open than Broomsgrove and has some undergrowth of brambles and St John's Wort (*Hypericum* sp.).

The proportion of unbanded and one-banded shells in these woods is the third highest, only less than in beechwoods and Rough Down. The general colour is pinkish-brown with a very much higher proportion of fawns than in the beechwoods. The proportion of yellows is the lowest of any habitat investigated.

The woods classed as mixed deciduous are a heterogeneous group. Wantage 1 and Streatley South Wood have a high proportion of beech, and the collections from them approach those from beechwoods in their character. Wytham and Bagley may be regarded as typical of these localities. Both are composed of a mixture of oak, ash, sycamore and other deciduous trees, with a certain amount of shrubs and with a plentiful undergrowth of brambles, bracken, grasses and woodland herbs. Bagley is the denser but neither is as dense as any of the oakwoods or beechwoods. Conifer plantations in Bagley were not visited. Burdrop Wood has a high proportion of ash and lies on an extremely steep Lower Chalk escarpment. Here, the collection was taken from an area containing much hazel and sloe with an undergrowth of brambles and nettles. Wantage 2 was a small open copse of elms on bare stony ground with young elm scrub and a little grass.

The proportion of unbanded and one-banded shells is much lower in mixed deciduous woods than in any other locality so far considered, but it is higher than in hedgerows and downland with coarse grass. The proportion of yellows is also high although less than in hedgerows and downsides. Yellows, fawns and pinks are approximately equally common.

The hedgerows are all principally of hawthorn with few and widely-spaced trees. All are bordered by rank green herbage, except the Parks Hedge which had been neglected and had a thick outgrowth of brambles, beneath which there was little green vegetation.

The average proportion of yellows in the hedgerows is higher than for any but downside localities (and it may be noted that the lowest proportion in any Hedge was found in the Parks Hedge collection). The proportion of unbanded and one-banded shells is the lowest except for downside with coarse grass, the difference between the two being very small.

The downside habitats all consist of herbage, sometimes with scattered bushes (usually hawthorn), on very thin soil above pure chalk, and all are on steep slopes. Christmas Common downside has scattered hawthorn bushes, and long coarse grass with a strong

mixture of rough herbs. The ground is broken in places, chiefly by the action of rabbits. The collection from the Pentridge (near Cranbourne, Dorset) was obtained through Dr D. A. Darcy, and the site has not been visited by us. From Dr Darcy's description it resembles Christmas Common downside strongly. It is a considerable distance from all the other stations, and may possibly be subject to slightly but significantly different climatic conditions. The proportion of unbanded and one-banded shells in these localities is the lowest observed, the proportion of yellows the highest.

Rough Down agrees with the other downsides in its general features, but there are no bushes near the collecting-area. The vegetation is extremely short cattle- and rabbit-grazed turf, and has been so for at least forty years. The ground is broken in many places through the action of moles, cattle, and rabbits on the very steep downside, and there are a number of chalk and flint stones, the former white, the latter dark brown or grey. The percentage of unbanded and one-banded shells is second only to that from beechwoods; the percentage of yellow second to that from downside with coarse grass.

The remaining two localities differ from those described above. The collection from East Ilsley was taken from among thick, long, dull-green, very uniform grass on a bank at the side of a downland track. The bases of the grass clumps were thickly clothed with dead leaves and stems so that the colour of the vegetation at the soil surface was uniformly brown. The shells were found in this layer, several showing evidence of predation by rats. The collection contained a high proportion of unbanded shells and a lower proportion of yellows. Nearly all the shells were brown. The collection from Sowberry Court was from a very neglected downland hedge that had widened and become almost a copse. There was very little undergrowth, the soil being covered with beech leaves and mast, larch cones and needles, and dead hawthorn leaves, all forming a dark brown carpet. There were many rabbit burrows. This collection had a very high proportion of brown shells and very few yellows. The proportion of unbanded shells was high; there were no one-banded shells.

DISCUSSION

It is obvious from tables 1 and 2 that there is a close relationship between the ratios of the colour and banding-patterns in any colony and the sort of background on which it lives. If one chooses the five localities with the most green vegetation at ground level, and the five with the most brown or red-brown carpet, the lowest percentage of yellow shells in any of the green habitats is 41 per cent. (114 in 279), the highest in the browns is only 17 per cent. (1 in 6), as shown in table 3. Intermediate habitats tend to intermediate values. To compare banding and discontinuity of background one must select

the five most uniform backgrounds, and the five showing greatest discontinuity. The results are shown in table 4. The lowest percentage of unbanded shells on a uniform background is 59 per cent. (94 in 159), the highest in variegated backgrounds is 22 per cent. (9 in 41). A general tendency to loss of bands is shown when one considers not only unbanded shells, but one-banded (00300) and those without

TABLE 3

Relation between greenness of background and percentage of yellow shells

Locality	Per cent. yellow shells	No. of shells in collection
(a) *Greenest*		
Wytham Lane	76	24
Pentridge	64	434
Rough Down	45	164
Christmas Common downside	43	158
Canal Bank	41	279
(b) *Least Green*		
Urchfont Hanger	17	6
Broomsgrove	8	97
Putall Gate	4	609
Aston Rowant	0	11
Shirburn Lodge	0	7

TABLE 4

Relation between uniformity of background and percentage of unbanded shells (00000)

Locality	Per cent. unbanded shells	No. of shells in collection
(a) *Most uniform*		
Aston Rowant	100·0	11
Rockley 1	74·0	156
East Ilsley	62·5	43
Rough Down	61·5	164
Hackpen	59·0	159
(b) *Least uniform*		
Hinksey Hedge	22·0	41
Wytham Lane	17·0	24
Christmas Common downside	12·0	158
Canal Bank	12·0	279
Parks Hedge	1·2	81

bands 1 and 2 as well. Bands 4 and 5 are not normally visible to any great extent and can vary without affecting the appearance of the living snail. Shells with such formulæ as 12300 can be classed with 12345 as effectively five-banded. The results of considering effective banding are shown in table 5. The lowest percentage of

effectively unbanded shells on uniform backgrounds is 79 per cent. (34 in 43) and the highest against a broken background is 39 per cent. (16 in 41).

TABLE 5

Relation between uniformity of background and percentage of effectively unbanded shells

Locality	Effectively 12345	Effectively 00000	Per cent. 00000
(a) *Most uniform*			
Aston Rowant	0	11	100
Rockley 1	0	156	100
East Ilsley	9	34	79
Rough Down	11	153	93
Hackpen	0	159	100
(b) *Least uniform*			
Hinksey Hedge	25	16	39
Wytham Lane	18	6	25
Christmas Common downside	102	56	35
Canal Bank	192	97	34
Parks Hedge	78	3	4

In fig. 3 is shown the relation between the percentage of effectively unbanded shells and the percentage of yellow shells for each locality. The connection between density of trees, high percentage of effectively unbanded shells, and low percentage of yellow shells, which corresponds to the relation between density of trees, uniformity of ground-litter and lack of green vegetation at ground level, is obvious. As the woods become more dense, their resemblances to each other in these respects increase, and the variation within the colonies of *Cepæa nemoralis* inhabiting them decreases correspondingly. Hedgerows appear as very reduced and very variable woods, those with most bramble being most like the rest of the woodland series, with respect to colour. Excepting that from East Ilsley, colonies from downside with long grass resemble those from the greener hedges. Rough Down is unlike all the others in that it is grazed. Consequently its vegetation is green but very short, and uniform. The colony on Rough Down is very similar in lack of banding to the beechwood and dense oakwood colonies.

It is our experience in collecting that heavily banded shells are more conspicuous than unbanded ones in uniform localities, whereas unbanded ones tend to be conspicuous in hedgerows and similar places. Also, if all the shells from a given locality are considered, it is seen that the general ground colour corresponds closely with the general colour of the background (except that hedgerow collections tend to be of yellows and pinks). For example, the proportion of fawns to pinks is much greater in unfelled oakwoods than in any beechwood. This close relationship is in fact much greater than is

shown by our simple classification into yellows, fawns, pinks, and so on, but is difficult to estimate colorimetrically.

The results obtained could be affected by two principal factors. First, we are, no doubt, not perfect collectors and the collections might have an undue proportion of shells appearing conspicuous to us. Secondly, in many localities quantities of shells collected originally by predators (thrushes, rats, grey squirrels, small field rodents, and rabbits) have been included in our collections. But

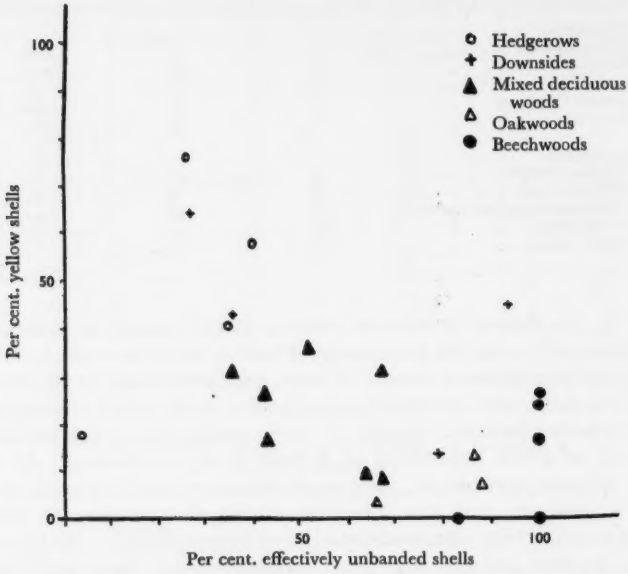

FIG. 3.—Relation between percentage of yellow shells and percentage of effectively unbanded shells in each locality. Note that localities classified together because of their ecological similarity (*e.g.* downland beechwoods or hedgerows) tend to group together. Sowberry Court has been included in the mixed deciduous woods, and East Ilsley in the downsides.

both these effects should, if operative, tend to obscure the correspondence which we have shown, and cannot increase it.

An explanation of the correspondence between background and colour-pattern of shell must be found. It has been stated (see, *e.g.* Taylor, 1907-14, p. 288) that shell coloration depends on the food-plants. But it has been observed by Lang (1904, 1912), Fisher and Diver (1934) and ourselves that colour is genetically controlled, and, further, that animals with shells of the most diverse colours, when all fed on the same food, *e.g.* lettuce, continue to add segments of the original colour to the growing shell. One does not find in nature shells showing numerous cross-bands of different colours. It is even

more difficult to explain the differences in banding on the grounds of difference in diet, and again, Lang (1904) and others have observed genetical control of the banding-patterns.

It has been suggested by Diver (1940) that differences between the colonies might well be due to genetical drift, along the lines indicated by Sewall Wright (*e.g.* 1940). But it is exceedingly improbable that if the colonies visited by us were all showing random divergence, all the beechwood colonies, for example, should have reached by chance such a remarkable similarity in stations often so many miles apart, especially while differing from those in other types of locality, which also show great similarities within the same locality-class.

The only other explanation which will account for this correspondence is natural selection acting upon genetic variation. *Cepæa nemoralis* is eminently polymorphic, so that one would expect a balance of different genetic forms in each population, maintained by the physiological advantage of certain gene-combinations but with the actual ratios of genotypes altered in different directions in different situations by the action of natural selection. Consequently the details of the results obtained in this survey cannot be assumed, without further investigation, to apply to colonies in widely distant parts of Britain, since differences in climate may well result in different physiological advantages of particular gene-combinations and so set up a different initial balance. (For example, unbanded pink shells might perhaps be more abundant in Cornwall because of a different physiological balance in that climate, but one would expect to find the relative proportions in different types of localities to be much as in the district investigated by us.) To determine the balance, the different localities chosen must be within a reasonably restricted area, otherwise an unknown part of the differences observed will be caused by changes in physiological advantage associated with difference in climate.

Although much work has been done on the genetics of *Cepæa nemoralis*, little of it appears to have been published. Haldane (1932) states that variations in the patterns are due either to multiple allelomorphy or to very closely linked sets of genes, but gives no evidence. From the work of Lang (1904, 8, 11, 12) and Fisher and Diver (1934) it appears that yellow is recessive to other colours, and the possession of bands is recessive to the lack of them. The results of Stelfox (1918) are open to the objection that he started with adults that may well have been previously fertilised. His F_2 generations point to linkage between colour and banding, but unfortunately the numbers given are too small to be significant. Fisher and Diver (1934) have shown linkage between colour and banding, apparently of varying degrees of closeness. From our study of the shells it appears that the 00300 condition is distinct from other bandings, and without intermediates, so that it is probably controlled by a

simple Mendelian mechanism. Intermediates can be found between most if not all of the other bandings. Breeding experiments are being undertaken to investigate these points.

In the course of our collecting, plentiful evidence has been obtained of predation by thrushes, grey squirrels, rats, small field rodents, and rabbits. No complete investigation has so far been made into the different intensities and kinds of selection by these animals. Experiments are in progress, but it should be pointed out that if selection is of the order of 1 per cent., which is considered large by Fisher (1930*b*), it may be almost impossible to demonstrate by normal sampling methods, but of course it may fluctuate considerably, and in consequence be easily detectable over short periods. It is also necessary to know the area from which the predators are collecting and the exact composition of the snail population within it, for which reason, as Robson and Richards (1936) have pointed out, the work of Trueman (1916) and Haviland and Pitt (1919) is inconclusive. The population can be estimated numerically by recapturing marked animals. We have already found a suitable marking technique for this purpose. The results obtained with it will also provide the necessary information on the distance which the predators carry the snails. Cott (1940) has shown that it is unnecessary to demonstrate that these predators have colour vision since tone is as important as colour.

Diver (1929) has shown from examination of fossil specimens that this species has been polymorphic at least since the Neolithic period. He concludes that the frequencies of different types of banding have remained approximately constant since then, but since the habitats occupied by his fossil specimens are unknown, this conclusion can be accepted only with the greatest reserve. It does not follow that other populations cœval with those of the Neolithic collection, for example, would also show the same banding-frequencies unless these were from the same sort of habitat.

Polymorphism in this species is therefore normal and stable. In every colony investigated by us (except one from which a sample of only 11 shells was obtained) at least two, and more usually five or more different phenotypes were observed, so that the various colonies are distinguished by ratios of phenotypes, not by the complete fixation of a single phenotype in each colony.

There are four principal types of polymorphism that could concern us here. Either an advantageous gene is spreading through a population (Ford, 1940, pp. 493-4), or there is balanced polymorphism with the heterozygote at an advantage (Fisher, 1930*b*), or particular gene-combinations are stabilised by linkage so that the recombination classes are infrequent (Haldane, 1930 ; Fisher, 1930*a* ; Ford, 1940), or one may have numerous populations showing random fluctuations as Sewall Wright suggests (*e.g.* 1940) and moving towards fixation of particular genes. The first and last possibilities are extremely

unlikely because the polymorphism appears stable, and the probability on either hypothesis of obtaining such a relationship as we have shown above is remote. Both the second and third agree with our results. Linkage has been shown by Fisher and Diver (1934) and by Stelfox as quoted by them.

In table 6 is shown the distribution of banding with respect to colour for most of our collections. The distribution of the different forms of banding is often not the same within the different colour

TABLE 6

(Only fresh shells were reckoned in compiling this table)

Locality	Yellow					Pink					Brown				
	00000	00300	12345	Others	Total	00000	00300	12345	Others	Total	00000	00300	12345	Others	Total
Downland Beech—															
Hackpen	15	24	...	...	39	76	39	...	...	115	1	1	...	...	2
Rockley 1	17	25	...	...	42	41	7	...	...	48	57	9	...	...	61
Oakwood—															
Broomsgrove	1	4	1	1	7	5	22	4	1	32	30	15	1	1	47
Cobham Frith	...	24	1	...	25	51	80	7	16	154	...	4	1	1	6
Putall Gate	9	6	4	3	22	135	165	62	138	500	59	...	...	...	59
Mixed Deciduous Wood—															
Streatley S. Wood	2	1	3	...	6	33	4	11	4	52	8	...	...	...	8
Wytham	1	7	33	28	69	15	15	54	40	124	24	...	...	6	30
Bagley	...	...	4	2	6	6	5	9	5	25	4	...	...	...	4
Burdrop Wood	1	4	18	14	37	17	3	8	10	38	23	1	...	...	24
Wantage 2	7	...	3	4	14	7	...	12	11	30	8	...	...	...	8
Hedgerows—															
Wytham Lane	2	1	10	3	16	...	...	2	2	4	...	...	...	1	1
Canal Bank	17	25	46	26	114	7	27	59	61	154	9	1	...	1	11
Hinksey Hedge	5	2	5	6	18	4	1	5	3	13	...	...	...	...	...
Parks Hedge	1	1	10	2	14	...	1	42	19	62	...	...	...	...	...
Downside, Long Coarse Grass—															
Pentridge	...	66	185	23	274	...	32	82	11	125	8	2	12	6	28
C. C. Downside	...	11	21	34	66	19	6	33	31	89	...	...	...	...	...
Downside Short Turf—															
Rough Down	47	21	...	6	74	5	13	...	7	25	49	11	...	5	65
Other Stations—															
E. Ilsley	...	...	6	...	6	...	1	...	...	1	27	4	2	2	35
Sowberry Court	1	...	2	4	7	12	...	5	14	31	32	...	...	...	32

classes. They are materially different in Rockley 1, Burdrop Wood, and Christmas Common downside, for example. (Banding in brown shells is usually extremely weak and difficult to determine except in some shells with formula 00300. It appears that there is interference by the colour factors in the expression of the banding-factors, so that brown shells must be disregarded for this purpose.)

Consider two pairs of alleles neither autosomally linked nor sex-linked. If one pair is in the ratio $p : q$, then in the absence of selection, in a very large population the ratio of genotypes in the next generation will be

$$p^2\,(\mathrm{AA}) : 2pq\,(\mathrm{A}a) : p^2\,(aa).$$

The situation is the same for the other pair. Let the ratio be

$$p_1^2\,(\mathrm{BB}) : 2p_1q_1\,(\mathrm{B}b) : q_1^2\,(bb).$$

If the progeny are classified by their constitution with respect to A and *a*, then as there is no linkage, the members of each class (AA or A*a* or *aa*) should be in the ratio

$$p_1^2\,(\mathrm{BB}) : 2p_1q_1\,(\mathrm{B}b) : q_1^2\,(bb).$$

Now if selection of any sort is acting, let the selective advantages of AA, A*a* and *aa* be respectively $x : y : z$, and for BB, etc. $x_1 : y_1 : z_1$. Now the ratios of genotypes in the next generation will be given by the expression

$$(xp^2\,(\mathrm{AA}) + y2pq\,(\mathrm{A}a) + zq^2\,(aa))\;\;(x_1p_1^2\,(\mathrm{BB}) + y_1 2p_1q_1\,(\mathrm{B}b) + z_1q_1^2\,(bb))$$

If these are now classed as phenotypes we have :

A and B	A and *b*	*a* and B	*a* and *b*
$xp^2x_1p_1^2$ (AABB)	$xp^2z_1q_1^2$ (AA*bb*)	$zq^2x_1p_1^2$ (*aa*BB)	$zq^2z_1q_1^2$ (*aabb*)
$2xp^2x_1p_1q_1$ (AABb)	$2ypqz_1q_1^2$ (A*abb*)	$2zq^2y_1p_1q_1$ (*aa*Bb)	...
$2ypqx_1p_1^2$ (A*a*BB)	...	...	...
$4ypqy_1p_1q_1$ (A*a*B*b*)	...	...	...

Whence it is easily seen that the ratio of B to *b* in class A is still the same as in class *a*. Consequently selection acting directly upon independent gene pairs cannot disturb this ratio. However, if there is interaction between the gene pairs (so that the selective advantage of any genotype is not simply derived from those of its genes) this situation will not hold. Physiological selection on genes only, not on genotypes, seems unlikely in these snails since the discrepancy between the ratios is in different directions in different colonies and is too large. Selection (by predation) of different phenotypes seems more likely. If so, since there will be continual recombination in each generation, all discrepancies must be produced by selection within the lifetime of each generation. In this case, selection must be fairly heavy, and easily detectable. But linkage may well be present. If so, and it is not very close, then the observed discrepancy will not be found without selection, since the less common classes will be increased by crossing over (from the commoner classes) more rapidly than they are decreased by it. If linkage is very close, the discrepancy might be expected, but our results do not suggest such a state of affairs.

With linkage and selection the discrepancy can be expected and will accumulate slowly until balanced by recombination. In this case, the amount of selection in each generation may be very small and difficult to detect. Our present collections are not large enough to allow any determination of the relative importance of physiological selection and selection by predators, but do show the importance of selection in general. Further evidence is being obtained from experiments involving breeding, and sampling natural and artificial populations.

Diver (1939, 1940) has shown that *Cepæa nemoralis* and *C. hortensis* are found in small breeding-communities, each containing considerably less than 100 individuals, within a colony. Particular patterns may be found only within one such community, or in a few close together. This non-random distribution he ascribes to genetical drift. Our observations in the field are in agreement with this explanation, but with the important proviso that the general facies of each colony is determined by selection, in part at least through the action of predators and in correlation with the background. Consequently, the degree to which any single breeding community can drift is limited. Within the range of our observations, drift cannot act against natural selection.

Cepæa nemoralis has been quoted by Haldane (1932), Robson and Richards (1936), Dobzhansky (1941), Huxley (1942), and Mayr (1942) on the evidence of Diver as giving an example of random distribution of varieties, the patterns of which have no selective value.

It is regrettable that most of the evidence from which widely quoted conclusions on this species were originally drawn remains unpublished and therefore inaccessible to zoologists in general.

Crampton's papers (1916, 1925, 1932) on the genus *Partula* are often quoted as giving evidence of random variation resulting from genetic drift. A close examination of them does not support this view. His very full data give good indications of clines in at least two species. Several species have extended their range ; this points to considerable migration. In one species, evidence is given that the proportions of certain varieties fluctuated markedly between the years 1906 and 1909. These fluctuations seem much too large to be caused by drift. Crampton's data are usually for whole valleys, sometimes for a third of a valley ; they are taken not from small isolated communities within a population, but from the population as a whole, which in many cases must be large. All these considerations tend to reduce the value of his data for suggesting the action of drift ; in fact, much of the evidence he produces suggests that natural selection is operating.

Welch's work (1938) on *Achatinella mustelina* Mighels is also widely quoted as a parallel. Welch gives no indication of the nature of the backgrounds on which this arboreal snail is found, nor of its predators. Looking through his illustrations, one gets the impression that this

species is remarkably variable, much as is *Cepæa*. His paper does not give the necessary data for conclusions on drift.

It seems that in view of the results presented above, all such cases of polymorphic species showing apparently random variation should be reinvestigated. No useful conclusions on this matter can be drawn merely from a knowledge of the distribution of varieties without a study of their habitats.

Diver (1940) has claimed that variations in shell colour and banding in the snail *Cepæa nemoralis* (L.) have no selective value, and occur at random in different colonies. He ascribes the differences between colonies to genetical drift.

These contentions cannot be sustained. There is a definite relationship between the proportions of different varieties in any colony, and the background on which they live. The more uniform the background (*e.g.* a continuous leaf-carpet, or an expanse of very closely grazed turf) the higher is the number of unbanded shells. Pink, fawn, red and brown shells vary in numbers and exact shade according to the general colour of the background. The proportion of yellow shells increases as the amount of green vegetation at ground level increases.

Thus there is good evidence that the general appearance of any colony is determined by natural selection. Predation is by thrushes, rats, squirrels, small field rodents, and rabbits. The close correspondence between the general appearance of each colony and its background suggests that predators hunting by sight are the agents of selection.

Polymorphism in this species is normal and stable, and is probably maintained by the physiological advantage of certain gene-combinations. The proportion of different forms of banding within the various colour classes are not always the same in any one colony. Consequently there must be either very close linkage between colour and banding, or linkage in some degree combined with selection for particular genes, or differential elimination of phenotypes, the last being probably the most important. Each colony is maintained in a state of stable polymorphism, but the proportions of the varieties are determined by natural selection. Whatever genetical drift may occur cannot act against natural selection, and is severely limited by it.

Cepæa nemoralis has been widely quoted as an example of a polymorphic species showing random distribution of non-adaptive genes. The work of Crampton on the genus *Partula* and of Welch on a species of *Achatinella* has also been quoted as giving evidence of genetical drift. The papers of these authors, however, provide no evidence for drift and in one instance give some evidence for selection. In view of this, all situations supposedly caused by drift should be reinvestigated.

SUMMARY

1. Collections of the land snail *Cepæa nemoralis* were taken from various localities in southern England.

2. The ground colour and band-formula of each shell was recorded.

3. Localities were classified into six main types according to their broad ecological features and the percentages of the different colour- and banding-varieties, found at localities in different ecological situations, were compared.

4. It was shown that there was a close relationship between the ratios of the colour and banding-patterns in any colony and the sort of background on which it lived.

5. Colonies of *C. nemoralis* living in the same type of ecological habitat tend to resemble one another, but those from different types of habitat tend to differ.

6. Selection, through the agency of predators hunting by sight, acting upon a stable polymorphic situation was shown to be the most important factor determining the general appearance of different populations.

7. Colour and banding patterns are genetically controlled and the species is stably polymorphic.

8. Linkage between colour and banding has been shown, but the difference in the distribution of banding-patterns in each colour class is probably the result of differential elimination of phenotypes.

9. It was suggested that all polymorphic situations, supposedly caused by drift, should be reinvestigated.

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POLYGENIC VARIABILITY IN CHROMOSOMES OF *DROSOPHILA MELANOGASTER* OBTAINED FROM THE WILD

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THE occurrence of heritable variation in wild populations of *Drosophila* species is now a commonplace. Flies which are trapped in the wild can be trusted to show genetic differences when suitably tested. Many of the variants to be found are traceable to single recessive mutant genes of large effect which are carried hidden in the heterozygous condition in the wild flies, revealing themselves when an inbreeding technique is used. These may be of the visible type, so called because they visibly affect some character of the fly, or they may be lethals or sublethals.

Other variation found in wild flies is less obviously traceable to single mutant genes, and indeed is almost certainly continuous in type and polygenic in inheritance. The variation in vigour and development found so plentifully by Dobzhansky in *D. pseudo-obscura* and *D. persimilis* is of this kind. Heritable continuous variation in the number of sternoplural chaetæ has been described in *D. melanogaster* by Wigan (1941). It seems likely that the large response to selection for increased chaeta number which Wigan obtained in his Ockley population is due to the spread of a single major mutant, but the response to low selection in the Ockley population and the responses to both high and low selection in the Ealing population seem to be genuinely due to changes in polygenic systems.

The present experiment was undertaken to pursue further the question of polygenic variation in wild populations and in particular to gain some idea of the amount of variability existing within chromosomes, but balanced in such a way that the genotype would contain more variability than was displayed in the phenotype.

THE METHOD OF EXPERIMENT

Attention was concentrated on chromosome II, since this was the easiest to manage with the stocks available. Fourteen flies were obtained by Mr L. G. Wigan from an Essex apple dump in 1946. Seven of the fourteen were found to carry lethals in their chromosome II's, one of them in fact having a lethal in each of its two chromosome

II's. An eighth fly was heterozygous for a sterility gene in its chromosome II (Wigan, 1948).

TABLE 1A

Data from the four homozygous lines

Generation	A/A		B/B (i)		B/B (ii)		C/C		D/D	
	H	L	H	L	H	L	H	L	H	L
F_1	40·15* *6·41*		36·80 *1·40*		31·15 *1·50*		40·98* *5·64*		43·46* *6·71*	
F_2	41·45 *5·90*		36·65 *5·30*		30·68† *0·65*		40·50 *5·00*		44·43* *6·03*	
S1	40·97* *6·07*	39·07* *6·72*	36·53 *3·94*	35·53 *2·35*	32·13 *1·65*	30·28 *−0·95*	40·08 *5·95*	39·30 *6·30*	failed	45·67* *5·78*
S2	40·88 *7·55*	40·65* *4·25*	33·90* *0·25*	37·30* *0·89*	31·13 *0·95*	31·65 *1·30*	42·05 *7·20*	41·13 *6·05*	... ...	43·34* *6·92*
S3	41·06* *5·32*	41·65 *8·30*	35·13 *3·15*	36·96* *1·78*	32·88 *2·95*	31·93 *2·35*	40·28 *5·75*	39·95 *6·40*	... ...	44·98* *1·20*
S4	40·28* *7·66*	40·91* *7·46*	36·88 *2·85*	34·95 *3·10*	30·93 *3·05*	30·45 *−0·50*	40·88 *6·35*	40·18 *6·25*	...	42·03† *−4·45*
S5	40·70† *7·10*	40·99* *4·63*	37·43 *2·95*	31·66* *2·47*	30·25 *1·75*	30·38 *1·65*	40·13 *6·75*	40·50 *4·70*	...	44·00* *4·00*
S6	38·84* *6·33*	39·95* *7·48*	37·65* *2·88*	32·55 *0·70*	31·10 *0·80*	30·80 *−1·20*	39·20 *5·80*	40·30 *6·10*	...	...
S7	failed	40·80 *6·90*	37·33* *4·44*	32·08 *1·25*	32·13* *0·41*	31·04* *2·07*	41·05 *6·90*	40·93 *6·05*	...	...
S8	...	40·62 *5·82*	37·40 *2·50*	32·80 *0·50*	30·25* *1·50*	31·25 *0·50*	40·78 *5·75*	40·18 *4·45*	...	...
S9	...	40·25 *4·60*	...	...	31·88 *0·35*	No counts	...	...	...	...
S10	...	...	...	...	32·90* *−0·60*	30·9 *1·60*	...	...	...	...
S11	...	...	...	...	32·90* *4·01*	31·97 *2·34*	...	...	...	...

The upper figure is the mean of the sex means
The lower figure is the difference of the sex means (female-male)
* Less than the full forty flies counted
† From a pool of the previous generation

A number of stocks were built up by Mr Wigan from these flies. Each stock was made homogenic for chromosomes X and III from the laboratory's standard Oregon inbred line: the stocks differed in chromosome II, each being deliberately made homozygous for a different chromosome II from the Essex population. No attempt was made to control chromosome IV.

TABLE 1B

Data from the six heterozygous lines

Generation	A/B		A/C		A/D		B/C		B/D		C/D	
	H	L	H	L	H	L	H	L	H	L	H	L
F_1	39·45 *5·30*		39·85 *6·10*		42·85* *3·59*		38·68 *6·45*		42·93 *2·85*		42·10 *5·90*	
F_2	39·00 *3·10*		40·13 *5·95*		42·70 *5·90*		39·28 *4·85*		42·53 *4·25*		42·23 *6·65*	
S1	39·73 *2·85*	40·70 *4·70*	38·75 *5·80*	39·58 *6·45*	44·15 *6·10*	41·93 *4·65*	39·38 *4·95*	37·05 *5·50*	42·75 *4·40*	40·73 *3·45*	41·70 *3·90*	40·38 *5·55*
S2	39·10 *6·30*	38·15 *6·10*	40·10 *6·40*	38·68 *5·35*	45·13 *7·55*	41·08* *7·04*	39·85 *5·50*	37·60 *3·80*	42·63 *1·55*	40·73 *4·05*	44·05 *6·20*	38·98 *3·65*
S3	40·30 *5·20*	37·55 *4·00*	41·58* *7·60*	39·08 *6·65*	44·88 *7·65*	42·00* *4·00*	40·38 *5·55*	37·70 *5·20*	43·88 *4·75*	42·23 *4·85*	42·45 *7·10*	38·93* *5·13*
S4	39·98 *2·35*	36·98 *3·75*	40·50† *8·80*	40·63 *6·35*	46·49* *8·11*	40·30† *5·80*	41·60 *6·60*	37·05 *4·80*	40·38 *5·05*	38·70 *5·20*	42·68 *7·65*	40·03 *5·55*
S5	40·63 *4·85*	38·16* *5·18*	...	39·05 *6·00*	48·68 *8·25*	37·49* *9·63*	41·35 *5·70*	37·15 *4·40*	40·08 *6·35*	38·18 *3·25*	41·60 *4·50*	38·70 *3·70*
S6	38·22* *6·44*	38·75* *3·50*	39·90† *6·60*	38·75 *5·70*	46·81* *7·72*	37·67* *9·33*	41·53 *5·05*	37·40 *2·10*	43·03 *4·55*	38·68* *6·25*	42·70 *5·60*	37·53 *5·45*
S7	41·12* *5·48*	36·18* *5·25*	43·15 *7·40*	39·03 *5·45*	49·07* *4·14*	39·73 *6·15*	42·13 *5·15*	38·30 *4·40*	45·55 *4·90*	37·98 *7·57*	43·10 *6·60*	37·81* *5·49*
S8	40·70* *7·40*	36·67* *4·04*	41·73 *8·25*	38·58 *5·55*	48·40* *8·80*	37·49* *5·82*	42·40 *5·30*	38·18 *6·25*	51·25* *6·69*	39·03 *5·25*	44·98 *3·45*	37·58 *5·45*
S9	39·48 *5·05*	36·00 *3·48*	41·73 *6·25*	39·50 *4·60*	...	...	41·25 *5·90*	37·68 *5·15*	...	36·93 *5·15*	...	...
S10	38·55 *5·40*	35·80 *5·10*	43·17* *8·04*	38·48 *5·15*	...	...	42·00 *5·20*	37·55 *3·20*	...	...	...	...
S11	37·75 *2·60*	35·80 *4·10*	41·43 *6·25*	39·48 *5·95*	...	...	42·50 *6·80*	38·23 *5·25*	...	...	...	...
S12	38·15 *1·70*	34·96* *0·93*	42·05 *6·90*	38·43 *5·05*	...	...	41·23 *5·65*	39·05 *5·70*	...	...	...	
S13	38·62* *1·63*	failed	40·94* *5·47*	37·23 *5·25*	...	...	41·68 *5·45*	38·18 *3·95*	...	...	...	...
S14	38·54* *4·73*	...	41·25 *6·15*	37·85 *6·40*	...	...	40·55 *5·90*	38·93 *4·05*	...	...	...	...
S15	40·41* *3·55*	...	42·27* *6·65*	38·15 *5·50*	...	...	42·30 *4·60*	38·35 *5·10*	...	...	...	...
S16	38·25* *5·00*	...	42·03 *4·95*	38·15 *6·10*	...	...	42·53 *6·25*	38·43 *3·45*	...	...	...	...
S17	...	...	41·67 *6·75*	35·52* *2·85*	...	...	41·43 *6·95*	36·92* *5·63*	...	...	...	...

For further particulars, see table 1A

Four of these stocks were kindly supplied to us by Mr Wigan after they had been maintained for a year in the laboratory. They were homozygous respectively for the chromosome II's which we will denote as A, B, C and D. These stocks and the lines to which they gave rise will be referred to as A/A, B/B, C/C and D/D. All the six possible crosses were made between these four stocks to give the lines which will be called correspondingly A/B, A/C, A/D, B/C, B/D and C/D.

Selection was practised for both increasing and decreasing numbers of abdominal chaetæ in all the ten lines, four homozygous and six heterozygous. The technique of the experiment differed slightly from that used by Mather and Harrison (1949). Each culture was the product of four parent flies, two females and two males, but only one culture was counted in each line in each generation. A second culture was raised but it was not used unless the first one gave too few flies for continuation of the line. Where available, twenty females and twenty males were taken for counting. The numbers of chaetæ were counted on the fourth and fifth abdominal segments and the counts pooled. The figures given in the tables are the means of the sex means of such pooled numbers, as were also used by Mather and Harrison (1949).

At times the first cultures failed to produce the desired twenty flies of each sex. These cases are denoted by an asterisk in table 1A, where the data are summarised. When very few flies were available, no attempt was made to select, and the unselected mass was pooled for use as parents of the next generation. These are marked by a dagger in the table. When both cultures failed completely, resort was made to the previous generation for continuation of the line.

Normally the two flies of each sex with the highest (in the high lines) or lowest (in the low lines) counts were taken for use as parents of the next generation. Thus with twenty flies of each sex, the selection would use 10 per cent. of the flies as effective parents. No selection was practised in the F_1's of the crosses, or in the corresponding generation in the four homozygous lines. In the F_2's, and in the corresponding generations of the homozygous lines, both high and low selections were made, so initiating the H and L lines which were subsequently kept distinct.

THE HOMOZYGOUS LINES

The data from the four homozygous lines are given in table 1A, and the results of selection are shown graphically in fig. 1.

The behaviour under selection of lines A/A and C/C follows the pattern expected from earlier experience of selection in inbred lines (Mather, 1941 ; Mather and Wigan, 1942) : no difference was established between the H and L lines up to S6 (*i.e.* the 6th generation of selection) in A/A and S8 in C/C when the selections ceased. The

L line of A/A was carried on for three generations after A/A H ceased at S6, and it continued to show no sign of change.

The first H selection from D/D failed and the line was not restarted. It is thus impossible to make any comparison between H and L in this line. It is clear from table 1A, however, that although the L line showed a slight downward drift there is no good evidence of it having a lower chaetæ count at S_5 than at the beginning of the experiment. There is thus no clear indication that selection was anything but ineffective as expected.

Line B/B behaved differently from the rest in that it showed an unexpected response to selection. This first appeared at S_4 and was complete at S_5 : it thus occupied only two generations. From S_5 to S8 no further change occurred. The change chiefly involved a response in the L line, and, when complete, led to the lines differing by about 5 chaetæ. This response to selection must imply genetic differences which might have arisen by mutation during the progress of selection, or, of course, might have been present in the original stock from which the selection lines were begun. This stock had been homogenic when it was first made, but as we have already seen, it had been kept for a year before the selection experiments began. Thus mutation might have occurred in the stock before selection. This was tested by extracting from the unselected stock a new homozygous line. The purification process was as follows.

$$\text{(stock)}\ \frac{B}{B} \times \frac{CyL^4}{Pm}\,;\,\frac{Sb}{Or}\ \text{(stock)}$$

$$\frac{CyL^4}{B}\,;\,\frac{Sb}{Or} \times \frac{CyL^4}{Pm}\,;\,\frac{Sb}{Or}$$

$$\frac{CyL^4}{B}\,;\,\frac{Or}{Or} \times \frac{CyL^4}{B}\,;\,\frac{Or}{Or}$$

$$\frac{B}{B}\,;\,\frac{Or}{Or}\ \text{(purified)}$$

Where *Or* indicates a chromosome from our inbred Oregon stock.

This new (second) homozygous B/B line was selected in both directions for 11 generations but failed to show any response. In chaeta number it corresponds to the L line of the original B/B selection from S5-S8 (table 1A and fig. 1).

The new B/B line showed no change under selection so that mutation cannot be a frequent and persistent phenomenon in the B/B stock. The L line of the original B/B was only extracted and stabilised after some generations of selection and yet it corresponds exactly to the second B/B line. Thus both the L line of first B/B and the whole of the second B/B must be presumed to have received the same genetic material and their only connection is via the original unselected stock. This genetic material was therefore present in

that stock. The early cultures of first B/B have, however, a higher chaeta number than either the L line derived from them or second B/B. This chaeta number was maintained and even increased slightly in H of the first B/B line. Thus the initial cultures of the first B/B, and also the original unselected line, must have contained chromosomes which gave higher chaeta numbers than those stabilised by the L selection. Now cultures are the product of four parents,

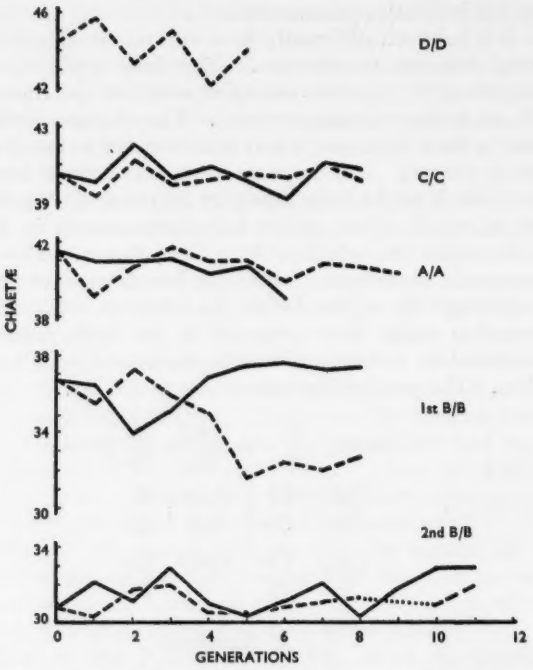

FIG. 1.—The effects of selection in the homozygous lines. Number of chaetæ (mean of sex means) is plotted against generations of selection. The H selections are shown as solid and the L selections as broken lines. A dotted line indicates the absence of observations for a generation.

2 females and 2 males, so that they contain 8 representatives of any chromosome. It is to be presumed that the sample of chromosomes with which first B/B started was mixed but included very few, perhaps only one, giving the low chaeta number. Thus it would take a few generations before the L line would be extracted and stabilised at its low level, which would depart from the initial average chaeta number by much more than the H selection would do. It will be observed that the data do not permit any certainty that the change was in chromosome II of the original stock. The purification process did not give a line certainly homozygous for chromosome X and III from

the B/B stock, although it might well have achieved this result in addition to the controlled homozygosis of B. If the second B/B line had responded to selection or if it had not corresponded to H or L of the first B/B line, some confidence could have been felt that chromosome X or III was involved in the change for it would then have been clear that the purification process, designed to deal with chromosome II, had not in fact been effective. Since, however, neither of these contingencies arose, uncertainty must persist as to whether the mutation was in chromosome II. In any case, as the original B/B line was used in the crosses with A/A, C/C and D/D, the interpretation of the results achieved by selection of the resulting three heterozygous lines must be somewhat in doubt.

Before passing on to consider the effect of selection on the heterozygous lines, certain differences between the homozygous lines must be observed. Table 2 shows the average numbers of chaetæ (mean of sex means) in all the lines, using families where at least 20 flies were available from the two sexes together. The H and L selections are pooled in A/A, C/C, D/D and the second B/B, where selection was ineffective. In the first B/B, the H and L lines are separated from S1 on. Since no effect of selection was apparent before S4, the inclusion of S1 to S3 in the separate lines must lead to a spuriously low value for the difference between them.

TABLE 2

Chaeta numbers in the homozygous lines

Line	A/A	C/C	D/D	First B/B		Second B/B
				H	L	
Number of families . .	14	18	4	8	8	23
Mean chaeta number . .	40·63 ±0·175	40·47 ±0·159	43·72 ±0·647	36·53 ±0·472	34·23 ±0·793	31·35 ±0·179

The standard errors of the means were found empirically from the variation observed between families within the line.

The mean numbers of chaetæ of A/A and C/C do not differ significantly ($t_{[30]} = 0{\cdot}671$) but all other comparisons among A/A, C/C, D/D and B/B show significant differences. The various chromosome II's thus differ in their effects on chaeta number. The difference between the H and L lines of the first B/B is barely significant on these data ($t_{[14]} = 2{\cdot}496$) and the difference between the second B/B and the L line of the first B/B is fully significant ($t_{[29]} = 3{\cdot}545$). These results are, however, open to objection because, as noted above, the H and L lines of the first B/B have been separated right from S1 in table 2, whereas no response to selection appeared before S4.

If we take only the data from S4 onwards in the first B/B, the mean numbers of chaetæ are H, 37·338, and L, 32·808. These differ significantly ($t_{[14]} = 3·883$) using the same estimates of error variation as in table 2. These estimates of error will, of course, themselves be too high since they will include the changes which have resulted from selection within the lines. The difference between the second B/B and the new value of the mean of L in the first B/B appears not significant ($t_{[9]} = 1·433$) but the significance may have been under-assessed as a result of the spuriously high estimate of error variation in the L line. The extent of the over-estimation is not, however, clear and there can be little doubt that no difference exists between second B/B and L of first B/B.

The various homozygous lines have been compared in respect of their sex differences, using only families in which at least 15 flies of each sex were available for comparison. D/D has been omitted from this comparison because it gave few families of the requisite size.

TABLE 3

Sex differences in the homozygous lines

Line	A/A	C/C	First B/B	Second B/B
Number of families . .	10	18	15	21
Sex difference . . .	6·77 ±0·274	5·97 ±0·204	2·43 ±0·341	1·13 ±0·288

The error variances were estimated empirically from the variation between the different families of each line. Those for A/A and C/C were found to be very similar and so were pooled to give a joint estimate of error variation of V = 0·750023 based on 26 degrees of freedom. The error variances of the two B/B lines (H and L being pooled in the first B/B) were similarly pooled to give a common estimate of V = 1·74891 based on 34 degrees of freedom. The standard errors of table 3 and the various tests of significance have been based on these pooled estimates of error variance.

There is a strong suggestion that A/A and C/C differ in their sex difference ($t_{[26]} = 2·359$, P = 0·05—0·02). We have already seen that the H and L selections of the first B/B show evidence of having departed in their sex difference. As expected the L line of first B/B does not differ significantly from the second B/B ($t_{[34]} = 0·379$). The contribution made by first B/B to the estimate of error variance will be inflated by the differences between the H and L lines that it contains ; but this inflation is hardly likely to be marking a difference between L of first B/B and second B/B when t as calculated is so small.

The differences between A/A and C/C and between the various B/B lines prohibit a simple comparison between A/A and C/C on

the one hand, and all the B/B lines on the other. The great disparity between the sex differences of the B/B lines and A/A and C/C can leave, however, little doubt that the B chromosome differs from the others in the sex difference it produces. It will be observed, too, that there is a difference in the variability of the sex difference. Even allowing for the inflation of the B/B error variation, the sex difference produced by the B chromosome seems definitely more variable than those of the A and C chromosomes.

One further comparison was made amongst the homozygous lines, viz. in their fertilities. This was measured very crudely by dividing the cultures into two classes : those that gave at least 20 flies of each sex and those that failed to do so. When a line failed (as opposed to being terminated deliberately) the failure was counted as one culture not giving 20 flies of each sex, and thereafter omitted from consideration. The results are given in table 4.

TABLE 4

Fertilities of the homozygous lines

Line	A/A	B/B		C/C	D/D
		1	2		
Cultures giving 20 flies of each sex .	6	12	17	12	0
Cultures not giving 20 flies of each sex	10	6	6	6	7
Total . . .	16	18	23	18	7

A test of homogeneity gives $\chi^2_{[4]} = 15{\cdot}844$ with $P = 0{\cdot}01 - 0{\cdot}001$. The main difference in fertility seems to lie between D/D and the rest. A/A and C/C show a suggestion of a difference in fertility but the evidence is not conclusive ($\chi^2_{[1]} = 2{\cdot}892$, with $P = 0{\cdot}10 - 0{\cdot}05$).

These various tests show that no two of the chromosome II's can be regarded as alike in their effects. Apart from A and C they differ in their effects on mean chaeta number, D giving the highest, B the lowest and A and C being intermediate. There are also differences in the sex differences in chaeta number produced by the chromosomes, and in this respect even A and C appear to differ. D gives flies of lower fertility than the rest of the chromosomes. Finally the lines fail, as expected, to respond to selection, apart from the difference which developed in the first B/B and which was traceable to its parent unselected stock.

THE HETEROZYGOUS LINES

As already noted, the four homozygous lines were intercrossed to give the six possible heterozygous lines, A/B, A/C, A/D, B/C, B/D and C/D. These were each subjected to selection for high (H) and

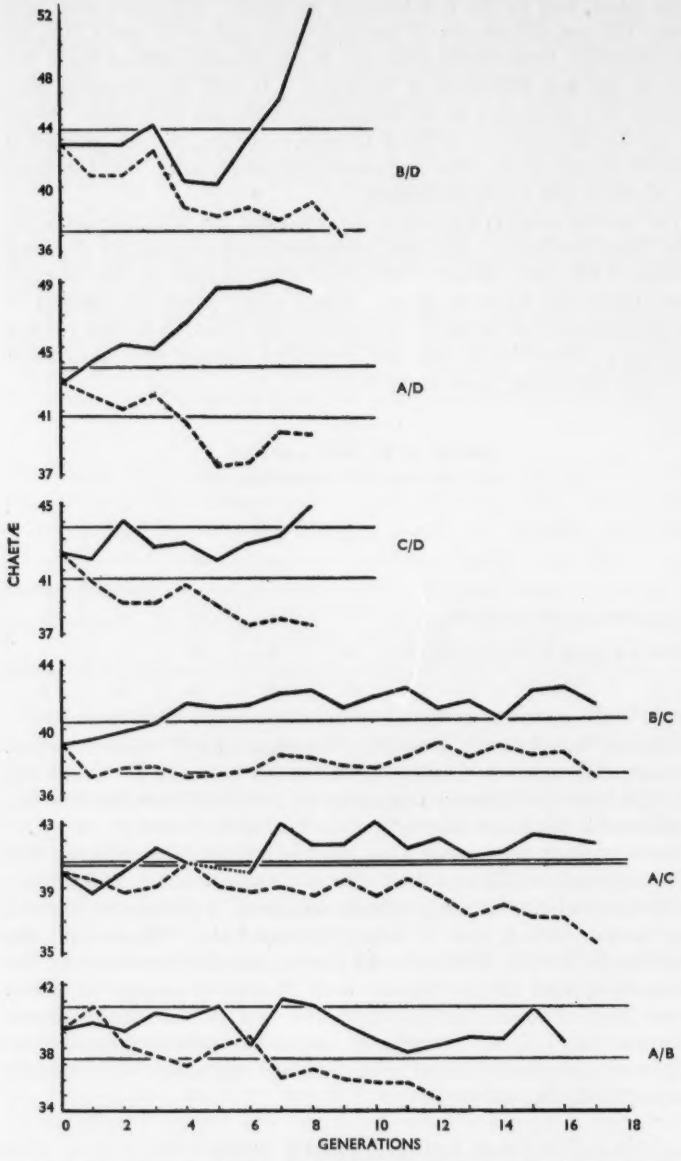

FIG. 2.—The effects of selection in the heterozygous lines. Number of chaetæ (mean of sex means) is plotted against generation of selection. The H selections are shown as solid and the L selections as broken lines. A dotted line indicated the absence of observations for a generation. The numbers of chaetæ of the parental lines (averages over all the generations shown in fig. 1) are indicated by thinner horizontal lines. The average of the H selection of first B/B (generations S4-S8) is used for the B/B line. The low level of this line, found as the average of second B/B, is 31·35.

low (L) numbers of abdominal chaetæ from F_2 onwards. The results of selection are given in table 1B and in fig. 2.

The mean chaeta numbers of the F_1 and F_2 generations were closely similar in each cross. Their average is compared for each cross with the chaeta numbers of the parental lines in table 5. Figures are given for both the H level (average of S4-S8 in H selection of 1st B/B) and, in brackets, the L level (average of 2nd B/B) in the case of the B/B parent.

TABLE 5

Comparison of the homozygous lines and their hybrids

Cross	A/B	A/C	A/D	B/C	B/D	C/D
Parents . .	40·63	40·63	40·63	37·34 (31·35)	37·34 (31·35)	40·47
	37·34 (31·35)	40·47	43·72	40·47	43·72	43·72
F_1 and F_2 .	39·23	39·99	42·78	39·98	42·73	42·17
Mid-parent .	38·99 (35·99)	40·55	42·18	38·91 (35·91)	40·53 (37·53)	42·10

In all cases, except A/C, the mean of F_1 and F_2 falls between the parental values, and lines A/A and C/C are so similar in their mean numbers of chaetæ that their F_1 and F_2 might well be expected to fall a little outside the parental range. In A/C, A/D and C/D the F_1 and F_2 average falls reasonably close to the mid-parent value, found as the average of the two parental means. In the three crosses which involve B, the F_1 and F_2 average is above the mid-parent even when the H value is taken for the B/B parent. The difference is not large in A/B when the H value is used for B/B ; but it is considerable in B/C and B/D. This suggests that chromosome B has a lower potency than C and D at least.

The responses to selection vary amongst the crosses (fig. 2). It shows the greatest combination of speed and size in A/D. Here both the parental limits had been transgressed by S4 ; and by S8, when the line was terminated, the difference between H and L was nearly 10 chaetæ, as compared with a difference of about 3 between the parents. Clearly there was a considerable amount of potential variability in this cross. It was released fairly smoothly and at a speed which made progress under selection over half as fast as that found by Mather and Harrison after crossing two distinct stocks, Oregon and Samarkand. Two chromosomes of the wild population carry, therefore, differences which can lead to remarkably rapid change under selection, these differences being largely balanced within the chromosomes.

Progress under selection in C/D is slower and smaller than in A/D. The difference between C/D and A/D is especially noticeable in the

failure of H from the former to pass the level of the upper parent before S8. Nevertheless, it would appear likely that H and L were still diverging when the line was terminated at S8, and a difference of some 6 chaetæ had been established between them by that time.

The divergence of H and L in B/D was very small before S6, but after that time it developed rapidly, largely because of progress by H, which at S7 had passed the upper parental level. The potential variability here seems to be no less than in A/D (in fact at S8 the difference between H and L of B/D is about 12 chaetæ), but it seems more difficult to release. Presumably the balancing genes are more tightly linked than in A/D.

It will be observed that the L selection of B/D failed to reach the lower parental value before S9, taking the H value as characteristic of B/B. Evidently the potential variability became more readily available to high than to low selection. This may, of course, have been due to the chance occurrence of some important but rare recombination earlier in the high line, than in the low line. It might, however, be due to the low recombinant chromosome being less viable, or at least less able to compete with its immediate parent chromosomes, than was its high counterpart. If we take the L value as characteristic of B/B, the L selection of B/D fails to reach the parental level, and these conclusions concerning the availability of potential variability are, of course, strengthened.

The A/B and B/C lines require little comment. H of B/C just exceeds the C/C level, and L of A/B just transgresses the B/B level (taking the H value for B/B) so suggesting a certain amount of potential variability in each cross. This conclusion is strengthened by the rather slow progress of both these selection lines in their early stages.

The homozygous lines A/A and C/C had mean chaeta numbers between which no significant difference could be detected. That chromosomes A and C were not, however, genetically equivalent was attested by a difference between the sex difference in chaeta number of the two lines. The genetical difference between the chromosomes is shown to extend to the genes affecting mean chaeta number itself by the results of selection in A/C. A difference between chaeta number in the H and L selections is clear after S7 and it seems to be increasing in magnitude right up to the S17 when the lines were terminated. By that time a difference of some 5 chaetæ had been established. Evidently, though displaying balances indistinguishable from one another, chromosomes A and C contained between them sufficient potential variability to permit marked, even if not large, advances under selection.

The foregoing observations refer to the mean chaeta number, found as the mean of the sex means. The sex difference in chaeta number in the heterozygous lines is also informative. The results, as they relate to the sex difference, are summarised in table 6.

As before the standard errors have been found empirically from the differences amongst the individual families in each line.

TABLE 6

Sex differences in chaeta number in the heterozygous lines

Line	A/B	A/C	A/D	B/C	B/D	C/D
H selection . .	4·15 ±0·37	6·65 ±0·29	7·56 ±0·59	5·68 ±0·25	4·78 ±0·46	5·73 ±0·39
L selection . .	4·46 ±0·44	5·54 ±0·27	5·60 ±0·72	4·58 ±0·25	4·86 ±0·43	5·05 ±0·39

The first point noticeable in this table is that the sex differences in the three lines one of whose parents was B/B are, with one exception, lower than those of the rest of the selections. The low sex difference of B/B is passed on to its offspring. The sex differences of the various lines descended from B/B are, however, all higher than the sex difference of B/B itself. If we compare them with the first B/B taken as a whole (*i.e.* pooling the H and L lines within B/B) all of the six lines (H and L from A/B, B/C and B/D) significantly exceed the parent in sex difference. If the comparison is made with the H line of first B/B the significance is lost, doubtless because only three families are available for the estimation of sex difference in H of the first B/B. The six heterozygous lines nevertheless all exceed H of first B/B in mean sex difference, even if not significantly, and we may feel some confidence that in fact none of the six had the genic constitution of B/B in this respect. This is not remarkable in the H lines of A/B, B/C and B/D ; but it is noteworthy in the corresponding L lines. It shows us that selection has not merely restored homozygosity for the B chromosomes in these L lines. There has been some recombination of the genes affecting the sex difference, recombination which may or may not have been effective in releasing potential variability in respect of the mean chaeta number.

The H line of A/B also has a sex difference significantly lower than that of A/A, this being doubtless related to the failure of this line to maintain consistently a level as high as the A/A parent in mean chaeta number. The H line of B/C is a little lower than that of C/C, but not significantly so. So far as sex difference goes, therefore, H from B/C could be carrying the C chromosome in homozygous condition. This is, however, unlikely as its mean chaeta number slightly exceeds that of C/C (fig. 2). The remaining sex differences are not informative.

The fertilities of the various heterozygous lines, found in the same way as used earlier for the homozygous lines, are given in table 7.

It is obvious from the table that the different lines differed in their fertilities. In general those lines one of whose parents was A/A have a lower fertility than those not descended from A/A. Only one

TABLE 7

Line	A/B		A/C		A/D		B/C		B/D		C/D	
	H	L	H	L	H	L	H	L	H	L	H	L
Cultures giving 20 flies of each sex	9	7	9	16	4	2	17	16	7	8	8	6
Cultures not giving 20 flies of each sex	7	5	8	1	4	6	0	1	1	1	0	2
Total . . .	16	12	17	17	8	8	17	17	8	9	8	8

comparison, however, requires special notice. The H and L lines of A/C differ significantly in their proportions of cultures which failed to give 20 flies of each sex ($\chi^2_{[1]} = 7{\cdot}404$). Thus the parental lines A/A and C/C, from which these two selections are descended, cannot have been genetically alike. This evidence reinforces the conclusion already reached from consideration of the sex differences in A/A and C/C, and of the effects of selection of the mean chaeta number in A/C.

THE GENETIC DIVERSITY OF THE FOUR CHROMOSOMES

It is clear from the observations described above that all the four second chromosomes, A, B, C and D, must differ in their genic content. B and D obviously differ from one another and from A and C in respect of their genes controlling mean chaeta number. Lines A/A and C/C give mean chaeta numbers which do not differ significantly; but they are not alike in their average sex differences in chaeta number. B also differs from both A and C in this latter respect. Few observations are available for D/D in respect of sex difference, but this line has a lower fertility than the rest.

The behaviour of the heterozygous lines under selection reinforces and extends the conclusions from observation of the homozygous lines. In interpreting the reactions of the heterozygous lines to selection, it is assumed that their changes are to be traced to selection of differences existing between the chromosomes of the parental lines, and that they are not due to selection of differences which have arisen *de novo* within the heterozygous lines after their origin by crossing. Previous experience had accorded with this view, it having been observed in particular that :—

(i) the speed and extent of advance under selection is the greater the more widely different, in respect of gene content, the parental

lines are expected from their origin to be (Mather, 1941 ; Sismanidis, 1942) ; (ii) the changes wrought by selection have a determinacy which would be difficult to understand if they depended on differences arising by mutation (Sismanidis, 1942 ; Mather and Harrison, 1949) ; (iii) change in homozygous lines, where selection must utilise variation arising *de novo*, is very slow (Mather, 1941 ; Mather and Wigan, 1942).

The present observations on the reaction to selection of the four homozygous lines adds further to our information in respect of point (iii). Lines A/A and C/C showed the expected absence of change. Line D/D was uninformative owing to failure of its H selection. Its L selection might be regarded as having fallen slightly in mean chaeta number ; but such an inference is unreliable in view of the absence of the corresponding H line and also in view of the low fertility of the L line itself. D/D could not be fairly regarded as contradicting other experience. Line B/B offers, however, a more serious objection. There is no doubt that its L selection fell. Furthermore a second, purified B/B line, which seemed to correspond to L of the first B/B, was stable under eleven generations of selection. This change in L of first B/B can, as we have seen, be attributed to mutation in the original stock ; but it does emphasise that our assumption, of change under selection in heterozygous lines being traceable to differences between the parents, is essentially statistical in its justification. Mutation does occur and can lead to change under selection. An individual case of change could be ascribed to mutation ; but the number, speed, size and determinacy of changes under selection in cross-bred lines are such that the vast majority of such changes cannot be ascribed to mutation. So we must assume that they are dependent on original differences between the parents.

It will therefore be assumed that selection in the heterozygous lines reveals differences between the chromosomes of the homozygous lines from which they are selected ; though a special caution is perhaps necessary in interpreting the changes in A/B, B/C and B/D. In general the evidence from these lines is most economically interpreted on the assumption that the usual B/B stock contributed its high rather than its low chromosome to them. The response to selection in A/C, interpreted on this basis, confirms the genic difference between chromosomes A and C, and indeed shows it to extend even to a character in respect of which the two chromosomes have indistinguishable overall balances. Their differences constitute potential as opposed to free variability in mean chaeta number. The existence of potential variability is also revealed by the behaviour of the three lines descended from D/D. In every case the H selection transcended the upper parent D/D, though only by a little in C/D. The low parent was passed by the L selection in A/D and C/D ; even if not in B/D. The occurrence of partially balanced differences between D and the other three chromosomes is thus clear, and what is more,

the difference established between H and L during eight generations in A/D indicates that this store of potential variability is greater than the free variation expressed as the observable differences between A/A and D/D, great though this latter is. In general, the differences between H and L selections are greater than the parental differences except in A/B and B/C. And, in these cases, as we have seen, selection did not re-extract chromosomes similar in genic content to both the parents, so that even here we have evidence of potential variability. Great as the phenotypic differences are between the homozygous lines, the genetic differences between their chromosomes are greater. Potential as well as free variability exists and is revealed by relatively short programmes of selection. There can be little doubt, when all the data are taken into account, that this small population of four chromosomes carried within it the genetic materials necessary for giving differences of 20 chaetæ or more, *i.e.* at least a 25 per cent. change of mean chaeta number in each direction.

The four chromosomes had been kept in the laboratory for a year between collection in the wild and testing. The question therefore arises as to how far the above conclusions can apply to the original wild chromosomes. There was no evidence of heterogeneity in A/A and C/C, and hence none of any change occurring in the laboratory. The evidence about D/D is much less complete but it at least gives no reliable indication of change. B/B undoubtedly did mutate during its period in the laboratory. We have, however, assumed above that it contributed to the heterozygous lines its high chaeta producing chromosome, *i.e.* the chromosome showing the lesser difference from A, C and D. There is thus no reason to doubt that the conclusions relating to polygenic differences among the four chromosomes used in the test would apply to the four chromosomes originally collected from the wild.

That the variation in mean chaeta number is under polygenic control becomes clear if we enquire into the minimum number of genes needed to explain the differences observed in this character. D/D and B/B differ from one another as well as from A/A and C/C. At least two genes must be postulated immediately. Now chromosomes A and C are not alike in genic content though alike in chaeta number. They must differ in at least two genes, the differences being balanced thus $+-$ and $-+$. D cannot differ from A and C merely by carrying the corresponding $++$ combination because the H and L selection of A/D transcend the D/D and A/A levels respectively and the L selection of C/D transcends the level of C/C. The difference between D on the one hand and A and C on the other requires therefore at least one gene additional to the two in which A and C are assumed to differ. A similar argument applies in respect of B. This cannot carry merely the $--$ combination of the genes distinguishing A and C, for the L selection from A/B seems not to be the same as B/B, and the H selection for B/C transcends the level of C/C. Thus at

least four genes are required to explain the differences among A, B, C and D. To these must be added the determinant which mutated in the original B/B stock, bringing the total up to five.

It would be easy to argue that there is evidence of the widening between the H and L lines from A/C being too prolonged to be fairly ascribable to a single recombination between two gene differences ; and that the same is true of C/D and B/D to name two others. In this way three further differences would be added. But even taking the bare minimum of five differences in these second chromosomes (assuming the change in B/B to be due to a gene in chromosome II), it is clear that the variation must be regarded as polygenic. It must be remembered, too, that Mather and Harrison (1949) found evidence of at least two gene differences affecting this character in chromosome X and at least three in chromosome III. Gene differences are, in fact, ubiquitous and it is pointless to argue whether there are only ten (which good fortune has enabled us to unearth so readily) or whether there are really a hundred of which the ten are a chance sample ; and whether the ten differences are in single genes or in compound effective factors (Mather, 1949). Finally, these differences are all unassociated with any known major mutations : they occur between flies all of which are wild type, even if some of their combinations would put the flies at a marked disadvantage in the wild.

SUMMARY

Four second chromosomes were extracted by breeding from flies taken on an apple dump in Essex. Four lines were made up, differing by being homozygous for these second chromosomes respectively, but being otherwise alike in having chromosomes X and III from the laboratory's standard inbred Oregon stock. The six heterozygous lines possible from crossing four such homozygous stocks were also made. The homozygous stocks had to be maintained unselected in the laboratory for a year before the experiments began.

Selection was practised in both homozygous and heterozygous lines for increased and decreased numbers of abdominal chaetæ.

No response to selection was observed in two of the homozygous lines. The data were incomplete for a third one, but here, too, no clear evidence of change under selection was obtained. The fourth line responded to low selection in the middle of the experiment but was stable subsequently. After purification from the original unselected stock the line also failed to show any further response. This response is therefore attributed to a non-recurrent mutation in the original stock, between its extraction and its use in the experiments. That the chromosomes distinguishing these four lines must all have been different genetically is shown by observations on mean numbers of abdominal chaetæ, sex difference in number of abdominal chaetæ, and fertility.

The responses to selection in the heterozygous lines confirm the genetical differences among the chromosomes in respect of chaeta number, and further show that much of the genetical variability was of the potential kind, being hidden in the form of balanced or partially balanced combinations of genes. It seems likely that the four chromosomes contained amongst them genetic differences sufficient to permit relatively rapid increase and decrease of the number of chaetæ by 25 per cent.

The chromosomes tested were separated by several generations from the wild ; but there is no indication of change between collection and experiment other than in one stock as already mentioned. Thus the chromosomes as tested may be fairly regarded as representing chromosomes collected from the wild.

At least five genes must be postulated in chromosome II to account for the differences and changes of chaeta number observed in the experiments. Taking earlier experience into account, at least ten genes must be postulated in the whole nucleus. It is very unlikely that the number of genes is in fact as low as this minimum.

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THE RELATION OF TAXONOMIC METHOD TO AN EXPLANATION OF ORGANIC EVOLUTION

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THE impact of Charles Darwin's classic, *The Origin of Species*, has been incalculable. The concept of evolution that he made explicit, and cogently documented, now permeates and gives direction to all biology. Yet, paradoxically, the orientation of his argument, directed as it was toward an explanation of the origin of species, has provided an impediment to study of the causal factors involved in evolution.

Species or specific types had been the focus of attention since the beginning of the Greek development of biology. Within the span of man's memory each kind seemed immutable, and their permanence, accorded religious sanction, had scarcely been challenged. In view of the evidence he had adduced, which could be adequately understood only in terms of an evolutionary hypothesis, it is not surprising that Darwin should have emphasised his refutation of the fixity of species. His monumental contribution, however, was his thesis that evolution proceeds from the natural selection of individual variants. Being handicapped by the lack of genetic evidence, he was forced to frame his argument in taxonomic terminology. He presented extensive data to show that characters which are individual in some instances are subspecific in others, and specific in still others. He also pointed to parallel variation in higher categories. He inferred from this evidence that specific and other typical character results ultimately from the particular differences of individual adaptedness.

Darwin's successors were no more able than he to explain the adaptive process involved in the formation of typical character. Imbued with taxonomic concepts, and impressed by the role of isolation in maintaining specific character for long periods, they regarded the species not only as a taxonomic category but also as an evolutionary unit. This view resulted from the failure to distinguish potential exchange of genetic materials within a species from the actual exchange which alone can be effective in evolution. As a consequence their efforts were directed toward description of the succession of organic types. The origin of a type was explained in terms of change of an earlier type rather than in terms of causally effective individual differences.

This taxonomic conception of evolution still persists. It persists, for example, in the use of categories, such as species, ecotypes, and biotypes, to denote the effective vehicles or so-called dynamic units

of evolution. It persists also in the use of the term " speciation " as a synonym of evolution, with the implication that the attainment of specific status is the cardinal point in evolution. The limits of species are defined by isolation ; but this term and the phrase " the origin of isolating mechanisms " are used ambiguously to denote not only isolation but also effects quite different, which operate far below the specific level. The issue is one of basic orientation. Acceptance to-day of Darwin's basic hypothesis requires the affirmation that evolution proceeds not from the similarities which constitute types and by which the members of categories are recognisable, but from the hereditary differences that adapt individuals to particular environments. This involves the postulate that in the living phase there exist two, and only two, effective vehicles of organic evolution : individuals and mating groups or breeding populations. This hypothesis can be tested only by a study of the adaptedness of individuals that actually interbreed in nature and the offspring they produce, regardless of typical character. Comparisons of species and the development of patterns of typical character provide no test. Taxonomic methods and concepts, which necessarily abstract typical character and overlook particular differences, can do no more than provide evidence that evolution has occurred. They can trace the pattern, but fail to reveal the effective factors and mechanisms concerned.

So far as a complex and multiform process can be characterised by one phrase, evolution can be said to be change of adaptedness. (This is not to imply, however, that change of adaptive character is a direct response to " need " for change.) Adaptedness is an attribute of individuals. It is a particular relational complex, the fitness of each individual in relation to its particular environment. It is genetic in origin, being determined by gene combination that results from the breeding of individuals sufficiently congruent with one another and with the external environment to survive and reproduce.

Adaptedness is not uniform for two reasons. First, individuals differ as a result of genetic variation, the basic cause of which is mutation. Second, the inorganic phase of the environment is highly varied : the earth's crust, the atmosphere, the sun's light and heat vary from place to place and from time to time. As Heraclitus long since observed, all is in flux. The result is a myriad of local facies of the total environment, a mosaic of factors variously combined and subject to continual change. If this mosaic is to be occupied by living organisms, an assortment of individuals variously appropriate in their adaptedness to these local facies is accordingly required ; as these facies change, the organisms adapted to them must change if the race is to persist.

The asexual organism requires for its persistence only one sort of adaptive response, namely, that to its external environment. It can

achieve change of adaptedness beyond its developmental plasticity (tolerance range, or adaptability, *sensu* Gause, 1942, 1947) only as a result of mutation. The evolutionary resources of clonal organisms are accordingly limited by the rate and character of mutations that occur, and these may be neither adequate nor appropriate to meet the conditions of environmental change. Any device that will permit the accumulation of a varied reserve of mutations without jeopardising survival of the individuals carrying them will prove advantageous to persistence of the group. For amongst such a store a genotype may occur that will be appropriate to a particular environmental change. This device is found in sexual reproduction, which provides for combination in one individual of alleles of different capacities, and hence for future individuals capable of different responses. Sexual reproduction therefore allows a new condition : a capacity of progeny to realise sorts of adaptedness different from those of their parents. This capacity for change of adaptedness of sexually reproducing organisms is adaptability. In this sense, adaptability is not an attribute of individuals, but rather a function of the reservoir of genes contributed by the individuals that actually interbreed. From this reserve are drawn, each breeding cycle, the genotypes that constitute a new generation and a new reserve.

Complete freedom of reproductive association would allow exploration of the totality of potential gene combination. This would have the conceivable advantage of providing individuals preadapted to all possible changes of environment. However, such unlimited combination would result in maladaptation of some or all of the progeny to the existing local environment, to which each reproducing individual is already adapted, and from which the group cannot ordinarily withdraw very far. A limit on the character of genotypes that contribute to a given reserve is accordingly required in order to prevent too great deviation of adaptive character in the progeny. To secure the range of genetic character required for adaptability to change, and at the same time to retain maximal adaptedness of progeny to existing conditions, a balance must be achieved between too much and too little combination, between the consequences of unlimited sexual reproduction on one hand and the adaptive limitations of asexual reproduction on the other. This balance, which is crucial to evolution, is effected through the agency of the breeding population, the limited group of individuals that actually, not only potentially, interbreed. The term breeding population not only denotes a group of reproductively associated individuals but also, by abstraction, connotes the limited and transitory gene reserve from which the genotypes embodied in the offspring are constituted. These interbreeding individuals are the material basis of evolution, and their reproductive association is its vehicle (see Darlington, 1939, 1940).

The breeding population, accordingly, is the effective evolutionary

reality, for it and only it can effect change of adaptedness in sexual organisms. The reproductive association and reciprocal interaction of its constituents, and the interaction of their progeny with the external environment, are the epitome of evolution, as Darwin perceived, and as Wright and Dobzhansky, amongst others, have so ably demonstrated. The reality of each breeding population is disclosed solely by its consequences, that is, by its progeny, and not by the possession of common attributes. Let it be remembered also that a breeding population can be drawn from individuals sufficiently divergent in external adaptive character as to be classified as different coexistent species. This is one meaning of introgressive hybridisation. Taxonomically, the breeding population has neither reality nor utility ; it is a genecological concept.

The causal importance of the mating group or breeding population was implied in Turesson's use of the word genecological and his original definition of the ecotype, but was not made explicit. On the contrary, it was obscured by his endeavour to express the interrelation between population and environment by means of typification and classification, rather than by consideration of the adaptive relations of the interbreeding individuals. Turesson originally defined the ecotype (1922, p. 102) as an " ecological unit to cover the product arising as a result of the genotypical response of an ecospecies to a particular habitat." Despite this taxonomic terminology, he at that time, as Gregor says (1947, p. 379), apparently regarded " his ecotype as a relatively homogeneous population separated from neighbouring ecotypes of different heredity by a narrow zone of intergradation." He ascribed the differences of heredity to environmental selection of appropriate genotypes. This was an auspicious beginning for an investigation of the causal factors of evolutionary process.

Conditioned, however, by the pervasive influence of taxonomic concepts in biology, that is, by the tendency to interpret particular relations in terms of types, Turesson apparently lost sight of Darwin's thesis that evolution proceeds from individual differences. He did not explore further the interrelations of each limited population with its effective environment, as would have been required in order to discover the adaptive factors operating in each case. Nor did he seek to analyse his " particular habitat " in terms of the response of particular genotypes. Instead, he attempted to classify these relations, to typify them. In this effort he necessarily abstracted the similarities of each population, for similarity, not difference, is the basis for assignment to any class. The ecotype accordingly became a type, as its name denotes, a taxonomic category characterised in this instance by physiological rather than morphological attributes. It became eventually the equivalent of the orthodox geographic race or subspecies (Turesson, 1925 ; Clausen, Keck and Hiesey, 1940). Like these categories the ecotype has been useful for reconnaissance, for description of the mosaic that the evolutionary processes have brought into being.

It has disclosed the fact that populations vary in their over-all adaptedness in accordance with general gradients of environmental difference. Its virtue is that in focusing attention on the physiological responses of subspecific populations, it has come closer to the factors that determine the adaptedness of genotypes. (The construction of this word is unfortunate. It is used, however, not in the sense of a type, but in the sense of the particular genetic constitution of an individual.) Nevertheless, neither analysis nor description in terms of similarity and typical response will suffice to disclose these factors. This will require other than taxonomic concepts and methods, with reference to both the organism and its habitat.

General comprehension of this requirement has been retarded by a tendency to confuse the taxonomic consequences of isolation with the restrictions on gene exchange that are requisite for adaptability. This confusion is more than a matter of words, as Dobzhansky (1940) has said of the ambiguous use of the term speciation. However, if the term isolation is limited to its literal sense of segregation, involving severance of reproductive relations, its distinction from restriction of gene exchange will be clarified. Isolation can then be employed to indicate specific status as suggested by Dobzhansky in his discussions of speciation (1940) and isolating mechanisms (1937). The agents of isolation, as he pointed out, are numerous and varied, and their effects may differ at different times. They may be elements of either the external or the reproductive environment. They may act independently and yet reinforce one another, producing jointly an effect that no one alone would be capable of. Dobzhansky uses the term speciation to denote this sort of effect. It is the stage in divergence at which restriction of gene exchange has become complete and final. This is the ground for the taxonomic utility of isolation. It is a permanent condition for the maintenance of types. It pertains not to individuals but to groups of individuals and categories. The effect of isolation on evolutionary process, however, is negative; as Dobzhansky has said, it "slows down evolution." Moreover, isolation may even be a prelude to extinction. For the suppression of intergroup gene exchange, with the attendant reduction of possible preadaptive combinations, will limit each isolate to the resources of its own gene reserves. The asymptote of isolation is the negation of sexual reproduction.

The term "isolating mechanisms" has been employed by Dobzhansky (1937) as "a convenient general name for all mechanisms *hindering* or *preventing* [italics added] the interbreeding of racial complexes or species." He distinguishes a number of types of agents within this general category of isolating mechanisms, and states (1941, p. 256): "It is an important fact, however, that any agent that hinders the interbreeding of groups of individuals produces the same genetic effect, namely, it diminishes or reduces to zero the frequency of the exchange of genes between the groups." This seems to imply

that the maintenance of some degree of restricted gene exchange, which may insure adaptability, has the same effect as suppression of gene exchange, which may destroy it. To avoid this confusion, we believe it essential to distinguish between diminished exchange, with its positive effects, and isolation, which tends to be negative with respect to adaptability.

In eliminating intergroup exchange, isolation closes an avenue that earlier provided the possibility of preadaptive combinations. The isolates are thereafter limited to the resources of their constituent and successive breeding populations : their genetic character and size, the reproductive relations already established amongst them, and their relations to the changing environment. Dobzhansky (1941, p. 255) pointed out that " If life is to endure, gene combinations whose adaptive value has been tested by natural selection must be preserved and protected from [the] disintegration " that would follow unlimited interbreeding. But this protection is secured primarily by the positive effects of agents that operate at a much lower level of organisation than the species. This is borne out by the fact that entities may become and remain sympatric only after gene exchange between them is suppressed sufficiently to allow each to maintain its specific character. Meanwhile, each may have become highly diversified, its parts being harmoniously related to the particular environments in which they occur. To state this concept more clearly and to avoid the equivocal use of the term isolation, we accordingly suggest the following paraphrase : " If life is to be perpetuated by sexual reproduction, a balance must be maintained by restriction of gene combination such as will engender both adaptedness of individuals and subsequent adaptedness of their progeny. This balance is effected by the association of individuals in limited breeding populations. The factors of the effective environment that determine the constituents of each population are to this extent determinants of evolution. The common adaptive characters by which a group is recognised are derived ultimately from those of particular breeding populations by intergroup gene exchange. The limits of these characters may be set by isolation."

The importance of isolation for the evolutionary process lies primarily in the fact that it permits greater diversification of adaptive character by setting limits to gene exchange. In this way it permits occupation of different effective environments in the same area by different genetic systems. It provides for a multiplicity of types. Dobzhansky has accordingly suggested that a premium is put on isolation. This follows logically from the widely accepted generalisation that sympatric species are reproductively isolated. However, this latter is true only of some. Evidence is accumulating to indicate that in many groups of organisms, populations stabilised in type, but still able to exchange genes, can coexist for long periods (for example : Hubbs, 1943 ; Wharton, 1944 ; Epling, 1947 ; Stebbins *et al.*, 1947 ;

Hovanitz, 1949); they are species even though they are not reproductively isolated. The evidence of their specific status lies in the fact that each maintains its identity despite reproductive contact with one another. Having preserved channels for gene exchange they may, on occasion, form new successful gene systems. These may be formed abruptly, as in the case of alloploidy, or by hybridisation without ploidy (Epling and Lewis, unpublished data) as Stebbins (1942) suggested might be the case; or gradually, as implied by Anderson's concept of introgressive hybridisation (1938). The evolutionary advantage inherent in this possibility of exchange between species is obvious. However, the effectiveness of such species resides not in their specific character nor in their "isolation," but in the character of certain of their constituent individuals which are able as a result of particular reproductive adaptedness to enter a breeding population and thus form new successful genotypes. Accordingly, we suggest that a premium more cogent to evolution is put on the maintenance of a restricted degree of gene exchange, even at the specific level (Epling, 1947). The fact that the genetic systems represented by some species can be preserved for long periods without reproductive isolation suggests that isolation may prove to be no more than a by-product of the reproductive adaptedness and mating character developed in certain groups (see Fisher, 1936). Isolation is not mandatory and may on occasion prove disadvantageous to evolution of the groups concerned.

The exchange of genes between individuals is conditioned in part by environmental factors that determine the distances over which transfer is effective. Related individuals are not uniformly spaced, but tend to be aggregated into communities of varied size and density. All the individuals of a given community do not necessarily exchange genes during one breeding cycle, and the community may accordingly be comprised of more than one effective breeding population. These may not be discrete, and in continuous large populations must be treated as statistical concepts. But, for convenience, these effective populations can be conceived of as discrete, and the exchange of genes between them in the course of time can be regarded as intergroup exchange. A species or its parts can accordingly be regarded as a system of breeding populations, the genetic integration of which varies because of the spatial position and different adaptive responses of its members. The character and extent of population diversity will vary in accordance with the degree of restriction imposed on gene exchange, that is, the amount of "gene flow" between its parts. The effect of intergroup gene exchange is to allow, but not require, adaptive character that arises in one part of a population to spread to another provided that it will enhance, or at least not impair, the environmental and reproductive adaptedness of the latter. As a consequence a given character may become typical of a population. Simpson (1944) has distinguished three styles, or modal aspects, of the evolution of types

which he has called speciation, quantum evolution and phyletic evolution. Should the character of intergroup exchange be limited so as to result in divergence between parts of a genetic system, such as might occur during great diversification of habitat, speciation in Simpson's sense might then ensue. Should a divergent element of a system be preadapted to an abrupt difference of effective environment, quantum evolution might be expected. Should intergroup gene exchange be facilitated throughout the system, the result might then be phyletic evolution. Recognition of these modes by the variant patterns of evolution that result from them is a function of taxonomic method ; but their recognition does not disclose the process and factors that have produced them. Whatever the mode or pattern, it is still the result of the constitution and particular relationships of each breeding population during this course of change.

Failure to comprehend the limitations of the taxonomic method is nowhere more vividly illustrated than by a recent statement by Goldschmidt (1948, p. 16) that palæontology is " the only science which actually sees parts of evolution *in action* " [italics added]. The methods of palæotaxonomy and neotaxonomy can do no more than abstract typical characters expressed in the phenotype. They delineate types which have arisen in nature. The homology of similar characters that appear in different types is usually assumed, but can be substantiated by genetic experiment in some instances. Having assumed or ascertained the homology of these characters, the taxonomist can then examine the frequencies with which they appear in similar groups. From the patterns thus determined, he can infer what the course of evolution may have been. One example will suffice.

The plant order *Malvales* is singularly homogeneous, that is, " natural." It is comprised of six families in which are included 223 genera (including *Malva*, *Gossypium* and *Hibiscus*, to which we shall refer) and more than 2000 species (Edlin, 1935). An ordinal character of considerable taxonomic importance is the leaf. Its total aspect in respect of shape and texture is usually diagnostic of the order and is so uniform that the leaves of one genus may readily be mistaken for those of another. To put it simply, an informed taxonomist looking at a leafy shoot can say with a considerable degree of certainty, " This appears to be one of the Mallows." The prevailing leaf shape in the order is ovate, with a tendency to be notched at the base, that is, heart-shaped. The venation is prevailingly palmate with the principal veins arising at the base of the leaf and diverging like the fingers of a hand. In pinnately veined leaves, a pair of basal veins is usually emphasised. A strong tendency also exists for the margin to be palmately lobed, usually obscurely or incompletely, with three lobes, but sometimes to the extent of producing five (or seven) well-defined lobes, or in some genera, even a digitate leaf with five leaflets.

The homology of this leaf character in different genera and families would doubtless be assumed by most taxonomists. In this instance, the

assumption has been shown to be well grounded. Stephens (1943, p. 315; 1945, p. 383) has shown that the same variants as those described above may occur within a species of cotton (*Gossypium*), some even on the same plant. In some species of cotton the leaves are all unlobed and heart-shaped. Stephens regards this as the juvenile type and has shown that in the course of development " all leaf shapes in the genus pass through, or are potentially capable of passing through three successive phases: (1) an entire (unlobed) leaf phase; (2) a phase in which the leaves at successive nodes become progressively lacinated into five lobes; (3) a climax phase in which leaf shape is stabilized " (1945, p. 380). The realised potential, and hence the climax, varies from species to species. This potentiality is genetically controlled, for the climax type that ultimately develops, whether unlobed, lobed, laciniate, or narrow and pinnately veined, is known to be dependent on a series of multiple factors allelic to one another between species (Hutchinson, 1934; Harland, 1939; Silow, 1939). The homology of genes and of typical character between species of *Gossypium* seems therefore clearly established, just as it has been between *Drosophila melanogaster* and *D. simulans* (Sturtevant, 1920, 1921). Furthermore, monogenic control of alternative leaf shape of a similar kind is known in the related genera *Malva* (Kristofferson, 1923) and *Hibiscus* (Deshpande, 1942), and Stephens (1945) has also shown that the developmental pattern of the leaf of *Hibiscus rosa-sinensis* is similar to that of *Gossypium*. By virtue of this similarity and probable homology between genera, the inference is drawn that this mode of leaf development had its inception in particular mutations, and that having become characteristic of the order, it now constitutes an ordinal type; or in other words, that types originate from particular differences. Evidences of this kind, provided chiefly by taxonomic methods, substantiated where possible by genetic methods, can be regarded as proofs of evolution; indeed, they are of the kind that Darwin relied on. But, although they indicate the general course evolution has followed, they do not illuminate the processes by which it has been accomplished.

Studies of breeding populations, on the other hand, and of the adaptedness and adaptability of the genotypes they represent, constitute an attack on evolution in process. They are genecological, not taxonomic. They inquire into the changing relations of genotype to genotype, and genotype to environment. They investigate particular causal relationships, not taxonomic types; they are a condition to understanding the ultimate formation of these types.

Formerly based primarily on observation and inference, this inquiry is now amenable to an experimental approach. It has scarcely begun. To prosecute it successfully requires a reorientation of physiology and ecology. The physiology of individuals considered alone, without cognisance of their genotypes, will no longer suffice, nor that of a group of cognate individuals whose relationships are postulated on no

more than specific resemblances. Nor will the ecology of associated species, which we call communities, suffice except to define the gross character of the habitat. The orientation of the future requires as its objective a study of the physiology and ecology of mating groups and the progeny that issues from them. Investigations in these fields will acquire meaning to the degree that the adaptedness and adaptability of the organisms concerned are taken as a guide.

It is necessary now to ascertain the mechanisms responsible for the assortment of genotypes in nature : their faculties of migration and the limits on establishment imposed by the character and rhythm of the particulars of the environment with which they may perchance react. This is to say that we must discover the mechanisms that determine what shall be the constituent individuals of successive breeding populations. For the variables that limit or increase reproductive and external adaptedness and the range of tolerance of both will determine the adaptive character of the breeding population and the adaptability of succeeding populations. This is the core of the evolutionary problem.

Two formidable technical barriers obstruct the approach to it : our limited knowledge of organisms whose genotypes, as encountered in nature, can be identified by appropriate markers ; and inadequate methods for studying the effective environment.

The organisms of the sort required, for example, are those whose genotypes can be identified and studied as to their reactions. A close approach to this requirement is known now in certain species of *Drosophila*. In this organism the chromosomal inversions, by suppression of recombination, tend to preserve the integrity of the genotype to which the chromosome in question contributes (Dobzhansky and Epling, 1948). At the same time, the inversion, and hence the genotype, so far as it is preserved intact by the inversion, can be identified by the visible banding of the salivary gland chromosomes. Thus two genotypes, somewhat approximate in constancy, but differing in adaptive values (Heuts, 1947 ; Epling *et al.* unpub.), can be combined and their interactions analysed. Advantageous and even unique as this material is, it is still no more than an approximation to the ideal. Whether more facile material can be found or constructed remains to be ascertained.

Assessment of the external environment is of equal importance, for what is being investigated is a complex particular relationship between the living and nonliving. To this end, attention must bear on the effective environment. By this we mean the particular external factors that, by their selective action on the genotypes presented to them, determine those that come to reproductive maturity and hence may become the constituents of any given breeding population. The effective environment is accordingly a conceptual complement of the effective breeding population. A simple example will illustrate our meaning. *Drosophila pseudoobscura* is active throughout the year in

the Sierran foothills, but the life span of an individual is measured in days. The seasonal changes in frequency of the carriers of different chromosomal types, which are known to differ in adaptive character, show clearly that, as the seasons progress, each succeeding population is subject to different daily rhythms, both physical and biotic (Dobzhansky, 1947). The adaptive relations that determine the constituents of one seasonal population do not determine those of another. Furthermore, certain species of a perennial *Delphinium* also occur in this region. Depending on the weather, these may remain dormant for several years, or at most may form a rosette of leaves. Flowering occurs only when a certain limiting threshold is passed. This threshold appears to be a physiological response subject to individual variation. Variation is further evidenced by the fact that the flowers of some individuals may wither before they have matured their seeds. The breeding population of a given year is accordingly determined by a particular association of variables rather than by "the environment" in a gross or constant sense. Even though *Drosophila* and *Delphinium* occur in the same locality, the fact is also obvious that the effective environment of one may bear little relation to that of the other.

The procedure for determining external environmental factors with present methods is primarily taxonomic, that is, to circumscribe a locality and ascertain the association of variables that characterises it. The adaptedness of the organism being investigated is then interpreted in terms of the environmental category thus created. Recognising that small differences of topography, for example, may modify such associations, the attempt is sometimes made to particularise them by reducing the locality to a "microenvironment," usually in a spatial sense. Although this device is initially useful in an exploratory way, the particular variables effective in determining the reproductive association of members of a breeding population and the survival of their progeny must still be investigated in particular causal terms. Tools are required that will somehow discriminate these relationships.

Similarities of phenotype may overlay differences of individual response that are crucial to change of adaptedness. Taxonomic methods and concepts can deal only with similarities of phenotype and must perforce overlook individual differences. Such methods, while adequate for recognition of the patterns already evolved, are inadequate for a study of evolutionary processes, for this must necessarily rest on knowledge of individual differences within the mating group. Accordingly, it is necessary now to recognise and reject the tendency that Gregor (1947, p. 390) has described as "the almost subconscious desire to resolve intra-specific variation into categories capable of being absorbed into the orthodox taxonomic system," such as subspecies, ecotypes and biotypes, and turn to an investigation of particular causal relations. The conceptual and

technical barriers are formidable. When they have been surmounted, then and only then, shall we begin to comprehend the processes that result in change of adaptedness.

SUMMARY

The orientation of evolutionary research is still predominantly taxonomic and concerned with the delineation of typical character, its development, and its subsequent localisation by reproductive isolation. These studies are essential to a proof of organic evolution. But, contrary to tacit or explicit assumptions often made, knowledge of typical character does not, and cannot, disclose the processes that effect change of adaptedness or the development of typical character itself. These processes can be disclosed only by study of the particular relations that exist between the individuals of an effective breeding population and its effective environment.

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THE INFLUENCE OF PARTIAL MANIFESTATION ON THE DETECTION OF LINKAGE

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I. INTRODUCTION

In the usual type of mating, involving two or more factors, where we can estimate the intensity of linkage from the proportion of recombinant types, there are a number of disturbing influences which prevent a simple interpretation of the data. The most well known are probably the occurrence of differential viability and the possibility of differential recombination fractions in male and female heterozygotes. The use of linkage backcrosses in both coupling and repulsion to overcome these difficulties is now a standard procedure. Another source of disturbance of the observed numbers is partial manifestation. Organisms, heterozygous for a dominant gene or homozygous for a recessive gene, may fail to exhibit the character in question in a proportion of cases and thus be effectively misclassified. Fairfield Smith (1937) considered the problem of estimating the degree of misclassification, but of much greater importance is the question how much are the usual *estimates of linkage* affected. We shall assume, for the most part, that differential viability is absent, as disturbances due to this cause are often small and require a large number of animals to reveal their presence, whereas partial manifestation may be quite considerable in fairly small bodies of data. There are two main methods of coping with this sort of complication : one can either try to set up matings showing full manifestation ; or one can develop the statistical analysis to deal with the difficulty. With experimental work at any rate one can employ both methods, although with human genetics only the latter course is open to us.

The basic problem is this : we have a character in which we are interested which appears to have imperfect penetrance—we have noticed skipped generations or a characteristic disturbance of the observed numbers—and we wish to examine the possibility of linkage to other genes, which may *themselves* not have complete manifestation. If linkage proves to be significant then we shall want to estimate it. A point of particular importance in the latter connection is the use of a method which will give an efficient estimate of the linkage when the degree of manifestation varies from one group of matings to another.

We shall be concerned mainly with methods applicable to the usual type of experimental data ; and shall conclude with a few illustrations of the treatment of population data such as are collected in the study of human genetics.

2. NOTE ON SCORING WITH SEVERAL PARAMETERS

Suppose the expectations involve several parameters : p ; θ_1, θ_2 . . . and we are particularly concerned with the significance and estimation of one of them, say p. Then if L is the logarithm of the likelihood, an efficient score for $p = p_1$ is $S(p_1) = \left.\frac{\partial L}{\partial p}\right|_{p=p_1}$, where the maximum likelihood estimates of the parameters : θ_1, θ_2 . . ., given $p = p_1$, have been substituted. It is easy to show that the amount of " partial " information about p is the reciprocal of the corresponding element in the inverse of the complete information matrix.

For large samples the appropriate test of significance is the χ^2 with one degree of freedom :

$$\chi^2 = \frac{S^2}{I_p} \quad . \quad . \quad . \quad . \quad . \quad (2.1)$$

where I_p is the partial information about p.

On the other hand we can find the maximum likelihood estimate of p by the usual method of interpolating between two sufficiently close scores of opposite sign. In this case the easiest way of calculating I_p is to use the formula

$$I_p \doteqdot \frac{S(p_1)-S(p_2)}{p_2-p_1} \quad . \quad . \quad . \quad . \quad (2.2)$$

where the score has been calculated at adjacent values, p_1 and p_2.

We have of course :—

$$\text{Var } p = I_p^{-1} \quad . \quad . \quad . \quad . \quad (2.3)$$

These results have been used to derive the tests of significance and the methods of estimation employed below.

3. LINKAGE BACKCROSS IN TWO FACTORS : *AB/ab = ab/ab*

Let us consider a linkage backcross with two factors in coupling of the type : AB/*ab* = *ab*/*ab* where A is misclassified as *a* in a proportion λ of cases. We can call the latter : A $\rightarrow$ *a*. We can use the formulæ given below for matings in repulsion and for misclassifications of the type, *a* $\rightarrow$ A, by a suitable transposition of the symbols. Let the observed numbers in the four phenotypic classes be :—

AB	A*b*	a*B*	*ab*	Total
a	*b*	*c*	*d*	*n*

The usual test for linkage in the absence of disturbing influences is :—

$$\chi^2 = \frac{\{(b+c)-(a+d)\}^2}{n} \quad . \quad . \quad . \quad (3.1)$$

If, however, we admit partial manifestation χ^2 is found to assume the modified form :—

$$\chi^2 = \frac{4(bc-ad)^2}{n(a+b)(c+d)} \quad . \quad . \quad . \quad . \quad (3.2)$$

We can clearly combine χ^2 obtained from families with different degrees of manifestation.

The maximum likelihood estimates for λ and p, the recombination fraction, are given by :—

$$\lambda = \frac{(cd-ab)}{(a+c)(b+d)} \quad . \quad . \quad . \quad . \quad (3.3)$$

$$p = \frac{b(a+c)}{b(a+c)+a(b+d)} \quad . \quad . \quad . \quad (3.4)$$

The variances of these estimates are :—

$$\text{var } \lambda = \frac{2(1-\lambda)}{n}\{2pq+\lambda(1-2pq)\} \quad . \quad . \quad . \quad (3.5)$$

$$\text{var } p = \frac{2pq}{n}\left(\frac{1}{1-\lambda}-2pq\right) = \frac{2pq}{n}(\theta-2pq) \quad . \quad . \quad (3.6)$$

$$\text{where } \theta = \frac{1}{1-\lambda}.$$

These formulæ assume that we can take λ as the same throughout the whole body of data. We shall consider below the problem of combining estimates from groups with different λ's.

Linkage matings will as a rule give more information about the existence of partial manifestation than those without linkage. In the present case the test for manifestation, admitting linkage, is :—

$$\chi^2 = \frac{4(ab-cd)^2}{n(a+d)(b+c)} \quad . \quad . \quad . \quad . \quad (3.7)$$

The amount of information about λ for small λ is n in a single backcross, but $\frac{n}{4pq}$ in a linkage backcross. There is a gain since $4pq < 1$ unless $p = q = \frac{1}{2}$.

For other types of mating, *i.e.* repulsion and $a \rightarrow$ A, we have the following changes. In each case the four classes are labelled (*a*), (*b*), (*c*) and (*d*) in turn.

TABLE 1

		(*a*)	(*b*)	(*c*)	(*d*)	
(1) AB/*ab* = *ab*/*ab*	; A → *a*	AB	A*b*	a*B*	*ab*	
(2) A*b*/a*B*	; A → *a*	A*b*	AB	*ab*	a*B*	p is estimated in each case.
(3) AB/*ab*	; *a* → A	*ab*	a*B*	A*b*	AB	
(4) A*b*/a*B*	; *a* → A	a*B*	*ab*	AB	A*b*	

Example

The data used here is taken from Professor Fisher's material involving the third chromosome of the house mouse. Pirouette, *pi*, is a simple recessive ; and light head, *te*, is a recessive in which the homozygote, *te*/*te*, does not always show the character. The matings are in coupling, *i.e.* $++/te\ pi = te\ pi/te\ pi$. This is clearly type (3) above. The observed numbers are :—

te pi	*te* +	+ *pi*	++	Total
40	16	93	102	251
a	*b*	*c*	*d*	*n*

From (3.2) we find $\chi^2 = 9{\cdot}82$, while (3.4) and (3.6) give $p = 31{\cdot}1 \pm 6{\cdot}6$ per cent.

4. ESTIMATE FROM THE MANIFESTING CLASSES ONLY

Before passing on to a more complicated case let us consider an approximation for p.

It is obvious that we can obtain a consistent estimate of the recombination fraction by considering the manifesting classes alone, *i.e.*

$$\check{p} = \frac{b}{a+b} \quad . \quad . \quad . \quad . \quad (4.1)$$

The expected number of individuals in the manifesting classes is $\frac{1}{2}n(1-\lambda)$ and so the variance of the above estimate is :—

$$\text{var}\ \check{p} = \frac{2pq}{n(1-\lambda)} \quad . \quad . \quad . \quad . \quad (4.2)$$

Comparing this with the variance of the estimate obtained when we employ all four classes, we can define the efficiency, E, as the ratio of the two quantities.

$$\text{Therefore } E = 1-2pq(1-\lambda) \quad . \quad . \quad . \quad (4.3)$$

It is easy to see that the efficiency increases with close linkage and low penetrance. For 10 per cent. recombination, $E = 0{\cdot}82$ for complete penetrance and is greater than $0{\cdot}91$ for $\lambda > 0{\cdot}5$.

5. COMBINING ESTIMATES OF p FROM DATA WITH VARYING DEGREES OF MANIFESTATION

This is a situation frequently met with in practice. Suppose the data falls naturally into groups with different degrees of manifestation. It is convenient to use :—

$$\theta = \frac{1}{1-\lambda} ; \ 1 \leqslant \theta \leqslant \infty \text{ for } 0 \leqslant \lambda \leqslant 1 \quad . \quad . \quad (5.1)$$

Applying the results given in section (2) above we find that for a *given group* the score for p is :—

$$S_p = -\frac{a}{q} + \frac{b}{p} + \frac{c}{\theta-q} - \frac{d}{\theta-p} \quad . \quad . \quad . \quad (5.2)$$

where θ is given by the quadratic :—

$$(a+b)\theta^2-(a+b+qc+pd)\theta+npq=0 \quad . \quad . \quad (5.3)$$

The amount of partial information about p is readily obtained from adjacent values of the score.

Example

Let us take another example from the third chromosome of the house mouse. This time we will consider backcrosses in coupling for luxate, *lx*, and dominant spotted, W. Luxate is here run as a dominant and often fails to manifest. Thus the matings are :—

$$lx\ \mathrm{W}/{+}{+} = {+}{+}/{+}{+}$$

i.e. type (1).

The observed numbers and the appropriate calculations are set out below :—

	(*a*) *lx*W	(*b*) *lx*+	(*c*) +W	(*d*) ++	Total
1.	24	7	65	69	165
2.	37	9	38	66	150
3.	19	2	12	30	63

	p	Quadratic for θ (>1)	θ	Score	I
1.	0·18	$31\theta^2-96\cdot72\theta+24\cdot3540=0$	2·84374	+15·8359	259·39
	0·19	$31\theta^2-96\cdot76\theta+25\cdot3935=0$	2·83205	+13·2420	
2.	0·18	$46\theta^2-89\cdot04\theta+22\cdot1400=0$	1·64265	+ 5·9467	386·54
	0·19	$46\theta^2-89\cdot32\theta+23\cdot0850=0$	1·63475	+ 2·0813	
3.	0·18	$21\theta^2-36\cdot24\theta+9\cdot2988=0$	1·41214	−16·1420	132·37
	0·19	$21\theta^2-36\cdot42\theta+9\cdot6957=0$	1·40588	−17·4657	

Summary

	Score $p=0\cdot18$	Score $p=0\cdot19$	I_p	$1-\lambda$
1.	+15·8359	+13·2420	259·39	0·35
2.	+ 5·9467	+ 2·0813	386·54	0·61
3.	−16·1420	−17·4657	132·37	0·71
	+ 5·6406	− 2·1424	778·30	

Interpolating gives $p=0\cdot1872$ and $(778\cdot30)^{-\frac{1}{2}}=0\cdot0358$.

Therefore $p=18\cdot72\pm3\cdot58$ per cent.

The estimate from the manifesting classes alone is 18/98, *i.e.* $p=18\cdot37$ per cent. The amount of information is now only 653·6, giving a standard error of 3·91 per cent.—this estimate has an efficiency of 84 per cent.

6. LINKAGE TO A PARTIALLY MANIFESTING CHARACTER: $AB/ab = ab/ab$; $A \to a$, $B \to b$

Suppose that B, instead of being a normal dominant character, also has incomplete manifestation of the type: $B \to b$, and let the proportion misclassified be ν. Then the test for linkage is :—

$$\chi^2 = \frac{n(bc-ad)^2}{(a+b)(a+c)(b+d)(c+d)} \quad . \quad . \quad . \quad (6.1)$$

This is immediately clear from the remark that if there is no linkage then the expected numbers are in the following proportions :—

$$(1-\lambda)(1-\nu) : (1-\lambda)(1+\nu) : (1+\lambda)(1-\nu) : (1+\lambda)(1+\nu)$$

and these can be rearranged in the form of a 2×2 contingency table.

The estimates of p, λ and ν are :—

$$p = 1 - \frac{an}{2(a+b)(a+c)} \; ; \; \lambda = \frac{(c+d)-(a+b)}{n} \; ; \; \nu = \frac{(b+d)-(a+c)}{n} \quad (6.2)$$

We also have :—

$$\text{var } p = an \frac{\{n(a^2+bc)-a(a+b)(a+c)\}}{4(a+b)^3(a+c)^3} \quad . \quad . \quad (6.3)$$

The various types of mating, with the appropriate modifications, are :—

TABLE 2

		(*a*)	(*b*)	(*c*)	(*d*)	Estimate
(1) $AB/ab = ab/ab$;	$A \to a$, $B \to b$	AB	A*b*	a*B*	*ab*	p
(2) A*b*/a*B*	$A \to a$, $B \to b$	AB	A*b*	a*B*	*ab*	q
(3) AB/ab	$a \to A$, $B \to b$	a*B*	*ab*	AB	A*b*	q
(4) A*b*/a*B*	$a \to A$, $B \to b$	a*B*	*ab*	AB	A*b*	p
(5) AB/ab	$a \to A$, $b \to B$	*ab*	a*B*	A*b*	AB	p
(6) A*b*/a*B*	$a \to A$, $b \to B$	*ab*	a*B*	A*b*	AB	q

With types (2), (3) and (6), (6.2) gives an estimate of q instead of p. The formulæ for λ and ν are unaffected.

Example

Let us make a direct estimate of the linkage between luxate and light head. The matings are : $+lx/te+ = te+/te+$; $te \to +$, $lx \to +$, *i.e.* type (3). The observed numbers are :—

te lx	*te* +	+ *lx*	++	Total
7	49	41	154	251
a	*b*	*c*	*d*	*n*

Applying the above results we obtain $\chi^2 = 2{\cdot}1$. This is not significant, but as the genes are known to be linked we can proceed to estimate the recombination fraction and, using (6.2) and (6.3), find : $p = 32{\cdot}7 \pm 10{\cdot}6$ per cent.

7. DIFFERENTIAL VIABILITY

Suppose we return to the case of A alone having partial manifestation, and wish to discover whether part of the discrepancy in the observed ratios is due to differential viability. Without a linked locus it is not possible to distinguish these two effects statistically. But if we can find a pair of linked genes, B and *b*, unaffected by partial manifestation or differential viability, then we are in a position to make the test, for the three degrees of freedom available between the four observed classes can now be allotted to linkage, manifestation and viability. We find :—

$$\chi^2 = \frac{\{(a+c)-(b+d)\}^2}{n} \quad . \quad . \quad . \quad (7.1)$$

which curiously enough is the χ^2 we should obtain if we were testing the segregation of B/*b* alone !

If differential viability is present then we can estimate p from the manifesting classes alone.

8. HUMAN FAMILIES

Now let us consider one or two examples of the treatment of data from human families. We shall use certain modifications of the method of *u*-statistics developed by Fisher (1935*a*, 1935*b* and 1936), and extended by Finney (1940) to cover a wide variety of instances. We will assume for simplicity that we have a test factor with dominance, represented by T/*t*, and that the frequencies of the allelomorphic genes for the test factor are unknown. In this case families with no *tt* children must be rejected. The analysis can be extended to the case where the frequencies are known in the manner described by Finney.

(*a*) "*Dominant*" *abnormalities*

Let us take the case of a so-called dominant abnormality, like diabetes insipidus, which sometimes skips generations and is assumed to be due to a single gene, D, with incomplete manifestation. The observed numbers are :—

DT	D*t*	*d*T	*dt*	Total
a	*b*	*c*	*d*	*n*

The use of Fisher's *u*-statistics gives the following familiar results for the scores and amounts of information with respect to a quantity $\zeta = 1-4\chi(1-\chi)$, where χ is the recombination fraction :—

(i) Double backcross, DT = *dt*.

$$\left.\begin{aligned} u_{11} &= (a-b-c+d)^2-(a+b+c+d) \\ \mathrm{S} &= \Sigma\tfrac{1}{2}u_{11}\,;\ \mathrm{I} = \Sigma\tfrac{1}{2}n(n-1) \end{aligned}\right\} \quad . \quad . \quad (8.1)$$

(ii) Single backcross (intercross for test factor), $DT = dT$.

$$\left.\begin{aligned} u_{31} &= (a-3b-c+3d)^2-(a+9b+c+9d) \\ S &= \Sigma\tfrac{1}{18}u_{31};\ I = \Sigma\tfrac{1}{18}n(n-1)\frac{4^n-3^{n-2}}{4^n-3^n} \end{aligned}\right\} \quad . \quad . \quad (8.2)$$

where Σ indicates summation over families.

Now suppose that a proportion λ of the abnormals are misclassified as normal. The appropriate modifications of the above results are, writing $\mu = \frac{1-\lambda}{1+\lambda}$:—

$$\left.\begin{aligned} &\text{(i) } DT = dt:\ D \to d. \\ &u'_{11} = (a-b-\mu c+\mu d)^2-(a+b+\mu^2 c+\mu^2 d) \\ &\quad = \{(a-b)^2-(a+b)\}-2\mu(a-b)(c-d) \\ &\qquad\qquad +\mu^2\{(c-d)^2-(c+d)\} \\ &S' = \Sigma\tfrac{1}{2}\mu'_{11};\ I' = \mu^2 I = \Sigma\tfrac{1}{2}\mu^2 n(n-1) \end{aligned}\right\} \quad . \quad . \quad (8.3)$$

$$\left.\begin{aligned} &\text{(ii) } DT = dT;\ D \to d. \\ &u'_{31} = (a-3b-\mu c+3\mu d)^2-(a+9b+\mu^2 c+9\mu^2 d) \\ &\quad = \{(a-3b)^2-(a+9b)\}-2\mu(a-3b)(c-3d) \\ &\qquad\qquad +\mu^2\{(c-3d)^2-(c+9d)\} \\ &S' = \Sigma\tfrac{1}{18}u'_{31};\ I' = \mu^2 I = \Sigma\tfrac{1}{18}\mu^2 n(n-1)\frac{4^n-3^{n-2}}{4^n-3^n} \end{aligned}\right\} \quad . \quad . \quad (8.4)$$

In each case the new score is a simple modification of the old and the amount of information is multiplied by a factor μ^2. μ is estimated by $\frac{\Sigma(a+b)}{\Sigma(c+d)}$ in both (i) and (ii). Thus in practice when we have data from both kinds of families we can estimate μ from the totals. If there is reason to suspect that μ differs in different sections of the data then the scores and amounts of information can be calculated for the various groups on the basis of the appropriate value of μ before combining. In the present case the scores are independent of the estimates of manifestation so we need consider only the u-statistics. It is worth noting that we shall be able to subsume under the present head families in which one parent, although not showing the character, is known from the pedigree to be heterozygous, provided of course that ascertainment is virtually through the parents. If ascertainment is through the children a slightly different treatment is required.

(*b*) *Recessive abnormalities*

Let us now consider a recessive defect in which the homozygous recessive sometimes fails to manifest, like the Laurence-Moon-Biedl syndrome. We will take two illustrations, both intercrosses with respect to the defect, one an intercross and one a backcross for the test factor. These cases are a little more complicated in that the

score for linkage is not now independent of manifestation and we are obliged to calculate the three information functions $I_{\zeta\zeta}$, $I_{\zeta\lambda}$ and $I_{\lambda\lambda}$. Ascertainment is assumed to be through the children. The observed numbers are :—

TR	Tr	tR	tr	Total
a	b	c	d	n

The modified results are as follows, where now $\mu = \frac{1-\lambda}{3+\lambda}$:—

(i) TR $= t$R ; $r \to$ R.

$$\tfrac{1}{3}u'_{31} = (\mu a-b-\mu c+d)^2-(\mu^2 a+b+\mu^2 c+d)$$
$$= \mu^2\{(a-c)^2-(a+c)\}-2\mu(a-c)(b-d)+\{(b-d)^2-(b+d)\}$$

where $\mu = \dfrac{\Sigma(b+d-1)}{\Sigma(a+c)}$.

$$S = \Sigma\tfrac{1}{18}u'_{31}.$$
$$I_{\zeta\zeta} = \Sigma\tfrac{1}{2}(n-1)\{(n-2)\mu^2+2\mu\}$$
$$I_{\lambda\lambda} = \Sigma\frac{(n-1)}{(1-\lambda)(3+\lambda)}$$
$$I_{\zeta\lambda} = \Sigma\frac{(n-1)}{(3+\lambda)(2^n-1)}$$

. . (8.5)

(ii) TR $=$ TR ; $r \to$ R.

$$\tfrac{1}{3}\mu'_{33} = (\mu a-b-3\mu c+3d)^2-(\mu^2 a+b+9\mu^2 c+9d)$$
$$= \mu^2\{(a-3c)^2-(a+9c)\}-2\mu(a-3c)(b-3d)+\{(b-3d)^2-(b+9d)\}$$

where $\mu = \dfrac{\Sigma(b+d-1)}{\Sigma(a+c)}$.

$$S = \Sigma\tfrac{1}{81}u'_{33}.$$
$$I_{\zeta\zeta} = \Sigma\tfrac{2}{9}(n-1)\{(n-2)\mu^2+2\mu\}\frac{4^n-3^{n-2}}{4^n-3^n}$$
$$I_{\lambda\lambda} = \Sigma\frac{(n-1)}{(1-\lambda)(3+\lambda)}$$
$$I_{\zeta\lambda} = \Sigma\frac{2.3^{n-2}(n-1)}{(3+\lambda)(4^n-3^n)}$$

. . (8.6)

The test for linkage, given partial manifestation, is :—

$$\chi^2 = S^2/I_\zeta$$
$$\text{where } I_\zeta = I_{\zeta\zeta}-\frac{I^2_{\zeta\lambda}}{I_{\lambda\lambda}}$$

. . . (8.7)

There are, of course, a very large number of different cases which might arise in practice and the above examples are merely intended to illustrate the general method of approach.

I am indebted to Professor R. A. Fisher for many useful discussions and for providing me with the data on the 3rd chromosome of the house mouse employed in the examples.

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NATURE GENETIQUE DES MUTANTS A DEFICIENCE RESPIRATOIRE DE LA SOUCHE B-II DE LA LEVURE DE BOULANGERIE

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I. INTRODUCTION

Toute population de levure de boulangerie (*Saccharomyces cerevisiæ*) contient un certain nombre de cellules mutantes (" petites ") qui, placées à la surface d'un milieu nutritif solide, donnent naissance à des colonies naines (Ephrussi, Hottinguer et Chimènes, 1949). La vitesse de croissance réduite de celles-ci est due à l'incapacité des levures mutantes d'utiliser le glucose par la voie respiratoire, de sorte que, même en aérobiose, elles fermentent le sucre au lieu de l'oxyder (Tavlitzki, 1949 ; Slonimski, 1949). La déficience respiratoire est, à son tour, la conséquence de l'absence de deux ferments respiratoires essentiels, la cytochrome-oxydase et la succino-déhydrase, fait établi à la fois par l'étude de l'activité enzymatique et par l'examen spectroscopique des cellules mutantes (Slonimski et Ephrussi, 1949).

La nature génétique de la mutation " petite colonie " a été étudiée sur la levure américaine " Yeast Foam " (Y.F.) de *S. cerevisiæ* (Ephrussi, Hottinguer et Tavlitzki, 1949). Les croisements entre une souche normale et une souche mutante, suivis de quatre backcross à la forme mutante, ont constamment donné naissance à des lignées normales ; le caractère mutant n'est réapparu que dans 5 spores sur un total de 596 spores étudiées. L'analyse de ces résultats a conduit à la conclusion qu'ils ne sont compatibles avec un mécanisme mendélien que si l'on admet que les formes normale et mutante diffèrent par un nombre élevé de gènes (>6 à 20). Mais une telle interprétation est en contradiction avec la fréquence élevée des mutations spontanées trouvée par ailleurs (Ephrussi, L'Héritier et Hottinguer, 1949). Il en a été déduit que la mutation est probablement de nature cytoplasmique. Enfin, l'ensemble des observations sur les mutants " petite colonie " et des données relatives au processus de mutation a conduit à l'hypothèse que les mutants en question diffèrent de la forme normale par la perte de particules cytoplasmiques autoreproductibles, essentielles à la synthèse d'un groupe d'enzymes respiratoires (*l.c.*).

Le mutant (1-A_2) de Yeast Foam dont l'étude vient d'être résumée est un mutant apparu au cours de la reproduction végétative de la levure. Des mutants de cette sorte apparaissent plus fréquemment encore dans une autre souche de levure de la même espèce, la souche

" Boulangerie II " (B-II), et tout porte à croire que le mécanisme de leur formation est analogue à celui révélé par l'étude des mutants de Yeast Foam, c'est-à-dire que la mutation porte sur un facteur cytoplasmique. Mais chez B-II on observe de plus que des mutants à propriétés analogues apparaissent souvent à la suite de la méiose : des ascopores isolés donnent fréquemment d'emblée des clones composés uniquement de cellules du type mutant. Les chiffres du tableau 1 permettent de constater que 44 pour cent des spores de

TABLEAU 1

Comparaison du taux de germination des ascopores et de la fréquence des spores mutantes dans les souches B-II et Y.F.

Souches	Nombre d'ascospores		
	isolées	germées	mutantes
B-II	596	27	12
Y.F.	112	85	4

B-II appartiennent à cette catégorie. Le taux de germination des spores de cette souche est si bas que nous n'avons jamais observé la germination des quatre spores d'un même asque et nous ignorons par conséquent le type de ségrégation qui s'y produit. Mais la fréquence totale des spores mutantes (44 pour cent) est suffisamment proche de 1/2 pour qu'il nous ait paru légitime de nous demander si le caractère mutant n'est pas contrôlé ici par un gène mendélien pour lequel la souche B-II serait hétérozygote, et si l'étude comparée des mutants prenant naissance dans les asques (" petites ségrégantes ") et des mutants apparaissant au cours de la reproduction végétative (" petites végétatives ") n'était pas susceptible de révéler l'existence d'interactions entre facteurs géniques et cytoplasmiques.

Le présent travail a été entrepris dans le dessein de répondre à ces questions.*

2. MATERIEL ET METHODES

(i) Toutes les *lignées de levures utilisées* dans ce travail ont pour origine deux souches de *Saccharomyces cerevisiæ* :

" Boulangerie II " qui provient de la collection du Laboratoire des Fermentations de l'Institut Pasteur de Paris ;

" Yeast Foam," levure de boulangerie américaine, que nous devons à l'obligeance du Professeur Ø. Winge.

Les souches B-15 et B-26 sont des lignées monosporiques haploïdes isolées à partir d'asques de B-II. B-15 est une " grande " de signe

* Une partie des résultats a fait l'objet de la thèse de Doctorat de S. Y. Chen (1950), présentée à l'Université de Paris.

(+), B-26 une " petite ségrégante " de signe (−). La souche B-15-p4 est une " petite végétative " isolée à partir de B-15 et est, comme elle, de signe (+).

La souche 1-A_2 est une " petite végétative " haploïde dérivée de Yeast Foam. Son origine a été décrite par Ephrussi, Hottinguer et Tavlitzki (1949). Nous rappellerons qu'elle est de signe (−) et qu'elle porte le gène *a* (exigence de l'adénine ; pigment rouge).

Toutes les autres souches qui seront mentionnées dérivent des croisements des précédentes.

(ii) *Les milieux de culture et de sporulation* sont les mêmes que ceux utilisés par Ephrussi, Hottinguer et Tavlitzki (1949). Il en est de même de la *technique des croisements.*

(iii) *Le diagnostic différentiel des " grandes " et des " petites "* peut être fait par trois méthodes : réaction colorée de l'indophénoloxydase (" Nadi "), détermination du QR, examen spectroscopique. Toutes les trois ont été appliquées à l'étude des souches mentionnées en (i). Par la suite, nous nous sommes limités au test du Nadi, plus simple et plus économique, effectué selon la méthode de Keilin (1929).

Dans ce travail, la réaction du Nadi nous servira à déterminer la présence de l'indophénoloxydase dans des clones monosporiques d'une part, et dans les cellules diploïdes formées à la suite de croisements de clones haploïdes, de l'autre. Cette deuxième utilisation appelle la remarque suivante. Lorsqu'on fait dans un tube un croisement en masse selon la méthode des Lindegren, ce tube contient au bout de 24 heures un mélange de cellules haploïdes, de figures de copulation et de cellules diploïdes. Les cellules diploïdes possédant un fort avantage sélectif sur les haploïdes, nous effectuons au moins 4 repiquages de la culture afin de l'enrichir en diploïdes et d'éliminer les haploïdes avant de la soumettre au test du Nadi. Cette technique est quelquefois remplacée par l'isolement direct, à l'aide du micro-manipulateur, de cellules diploïdes.

La réaction du Nadi est effectuée macroscopiquement sur la suspension de levure lavée.

3. ANALYSE GENETIQUE DES MUTANTS " PETITE COLONIE "

(a) Démonstration d'une différence génétique entre " petites végétatives " et " petites ségrégantes." Réapparition de la forme normale dans les croisements des deux types de " petites."

L'existence d'une différence génétique entre " petites végétatives " et " petites ségrégantes " est immédiatement révélée par le croisement de ces deux formes avec une même levure normale.

Le croisement (1) de la " petite végétative " 1-A_2 (portant le gène *a*) avec la " grande " B-15 (qui porte le gène *A*) fournit des cultures diploïdes Nadi positives. 14 asques formés par celles-ci ont été analysés. Ils contenaient tous 2 spores *A* et 2 spores *a*. D'un autre côté, toutes les 56 spores ont donné naissance à des clones Nadi positifs.

Ce croisement fournit donc une F_1 entièrement constituée par des " grandes." Le caractère mutant disparaît ici comme dans les croisements étudiés précédemment (*l.c.*).

Le croisement (53) de la même " grande " B-15 avec la " petite ségrégante " B-26 fournit un résultat différent. Les diploïdes hybrides sont encore une fois Nadi positifs, mais, dans ce cas, le caractère mutant réapparait en F_1. Les 32 asques analysés ont montré une ségrégation régulière en 2 " grandes " : 2 " petites."

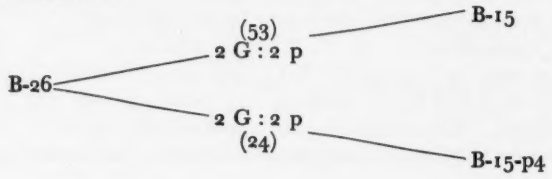
Ainsi, les croisements d'une même " grande " avec deux " petites " différentes, l'une " végétative," l'autre " ségrégante," fournissent des résultats différents. Dans le premier cas, le caractère mutant ne réapparait pas dans les asques de la F_1. Le second croisement est, au contraire, suivi d'une ségrégation mendélienne régulière. Il en ressort qu'un même phénotype mutant peut reposer sur des bases génétiques différentes. Chez la " petite ségrégante," le caractère mutant est sous la dépendance d'un gène mendélien.

La comparaison qui vient d'être faite porte cependant sur deux " petites " qui diffèrent en outre par la souche d'origine. Afin de la reproduire sans sortir des limites d'une souche, nous avons isolé, à partir de la souche B-15, la " petite végétative " B-15-p4. Le résultat du croisement (53) entre une " grande " et une " petite ségrégante " de la souche " Boulangerie II " étant déjà établi, et le croisement entre la même " grande " B-15 et la " petite végétative " B-15-p4 étant impossible (puisque ces deux souches sont du même " signe "), nous avons effectué le seul croisement possible (24) : " petite ségrégante " B-26 × " petite végétative " B-15-p4. La culture diploïde obtenue était Nadi positive et l'analyse de 15 asques a montré qu'ils contenaient tous 2 " grandes " et 2 " petites."

Cette expérience apporte, on le voit, un résultat nouveau : le croisement de deux " petites " fournit des asques dans lesquels on observe régulièrement une ségrégation simple en 2 " grandes " : 2 " petites." Ce résultat confirme d'une façon indiscutable la conclusion précédente quant à la différence génétique entre " petites végétatives " et " petites ségrégantes." En outre, elle apporte une justification à la désignation par des termes différents de ces deux sortes de mutants qui présentent le même phénotype et ne diffèrent que par leur mode d'origine.

(b) Essai d'interprétation mendélienne des résultats précédents

Les résultats qui viennent d'être décrits peuvent être représentés par le schéma suivant :



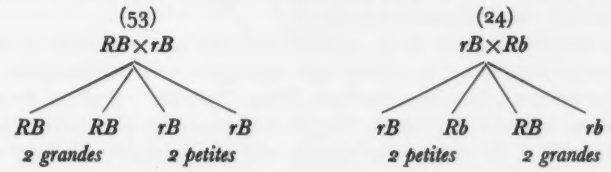
qui met en évidence des relations à première vue paradoxales et dont l'interprétation en termes mendéliens s'avère difficile.

Le croisement 53 conduit à une ségrégation monofactorielle typique : 2 " grandes " : 2 " petites." Comme l'examen des cellules diploïdes qui donnent naissance à ces asques montre que le caractère normal est dominant (les cultures diploïdes sont Nadi positives), nous pouvons en conclure que la souche B-15 porte un gène dominant (*R*) et la souche B-26 son allélomorphe récessif (*r*). D'un autre côté, le croisement (24) conduit également à la formation de diploïdes Nadi positifs et d'asques à ségrégation en 2 " grandes " : 2 " petites." Le type de ségrégation semble donc indiquer que les souches B-26 et B-15-p4 diffèrent aussi par un seul gène, mais ceci est incompatible avec le fait qu'ici le croisement de deux " petites " redonne des formes normales. La réapparition de celles-ci exige, semble-t-il, l'intervention d'au moins deux gènes non allélomorphes.

On peut cependant admettre que la ségrégation 2 : 2 observée dans ce dernier cas n'est monofactorielle qu'en apparence et que nous sommes en présence d'une interaction de deux gènes telle que le phénotype normal est déterminé aussi bien par la présence simultanée de deux dominants que par celle de deux récessifs non allélomorphes. Supposons, comme précédemment, que la " petite ségrégante " B-26 diffère de la " grande " B-15 par un seul gène, dont elle porte la forme récessive *r*. Supposons, d'autre part, que la souche " petite végétative " B-15-p4 résulte de la mutation récessive à un autre locus, soit *B*. Les trois souches étudiées ont alors les génotypes :

Grande	B-15	*RB*
Petite ségrégante . . .	B-26	*rB*
Petite végétative . . .	B-15-p4	*Rb*

Admettons, enfin, que la méïose dans les hybrides comporte un crossing-over obligatoire entre les loci *R* et *B*. Les résultats des croisements 53 et 24 peuvent alors être représentés comme suit :



Quoique ce schéma nous ait paru peu vraisemblable, nous l'avons soumis à la vérification expérimentale, car c'est le seul schéma mendélien qui nous ait paru susceptible de rendre compte des observations décrites.

La vérification repose sur le raisonnement suivant : selon le schéma ci-dessus, les asques hybrides du croisement 24 contiennent obligatoirement deux spores " grandes " dont les génotypes sont *RB* et *rb*, et deux " petites " l'une *rB*, l'autre *Rb*. Aussi bien les deux " grandes "

que les deux " petites " auraient donc des génotypes " complémentaires." Les croisements entre deux " petites " ou deux " grandes " *d'un même asque* devraient par conséquent conduire :

(i) à des diploïdes " Nadi " positifs
(ii) à des asques montrant une ségrégation 2 " grandes " : 2 " petites."

L'asque No 13 du croisement 24 nous a permis d'entreprendre cette expérience : ses spores *b* et *d* sont des " grandes " de signe opposé et *a* et *c* des " petites " de signe opposé. Nous avons donc réalisé les croisements entre les deux " grandes " (63) et entre les deux " petites " (66).

Les résultats du croisement 63 sont les suivants : l'hybride est Nadi positif. Les asques contiennent tous 4 spores normales.

D'un autre côté, l'hybride du croisement 66 est Nadi négatif et ne sporule pas.

Ni la ségrégation observée à la suite du croisement 63, ni l'absence d'indophénoloxydase dans les cellules diploïdes formées dans le croisement 66 ne sont compatibles avec l'hypothèse que nous avons cherché à vérifier.

(c) *Hypothèse invoquant un facteur cytoplasmique*

L'interprétation, en termes mendéliens, des résultats des croisements 24 et 53 s'étant avérée difficile sinon impossible, nous avons formulé une autre hypothèse que nous allons exposer maintenant.

Supposons que la " petite végétative " B-15-p4 possède le même génotype (*R*) que la souche B-15 dont elle dérive et résulte de la perte d'un facteur cytoplasmique essentiel à la synthèse des ferments respiratoires. Supposons d'autre part, que ce facteur cytoplasmique, autonome dans sa reproduction, ne fonctionne normalement qu'en présence du gène dominant *R*. Il suffit alors d'admettre que la " petite ségrégante " B-26 porte le gène *r*, pour expliquer l'ensemble des résultats des croisements 53 et 24 :

Les souches B-15 et B-26 ne différant que par le couple de gènes *R*/*r*, leur croisement doit fournir une ségrégation monofactorielle.

D'autre part, le croisement des deux " petites " B-26 et B-15-p4, dont l'une apporte le gène *R*, l'autre le facteur cytoplasmique inactif, doit aboutir à la reconstitution du système complet (facteur cytoplasmique actif en présence de *R*) et à la réapparition de spores à phénotype " grande " dans les asques hybrides, qui montreront ainsi une ségrégation monofactorielle.

Cette interprétation rend donc parfaitement compte des résultats des croisements 53 et 24, et il est facile de voir qu'elle est également en accord avec ceux des croisements 63 et 66. En effet, selon cette nouvelle interprétation, les génotypes des deux " petites " de l'asque No 13 d'une part, ceux des deux " grandes " du même asque, d'autre part, sont pareils ; les deux premières portent le gène *r*, les deux

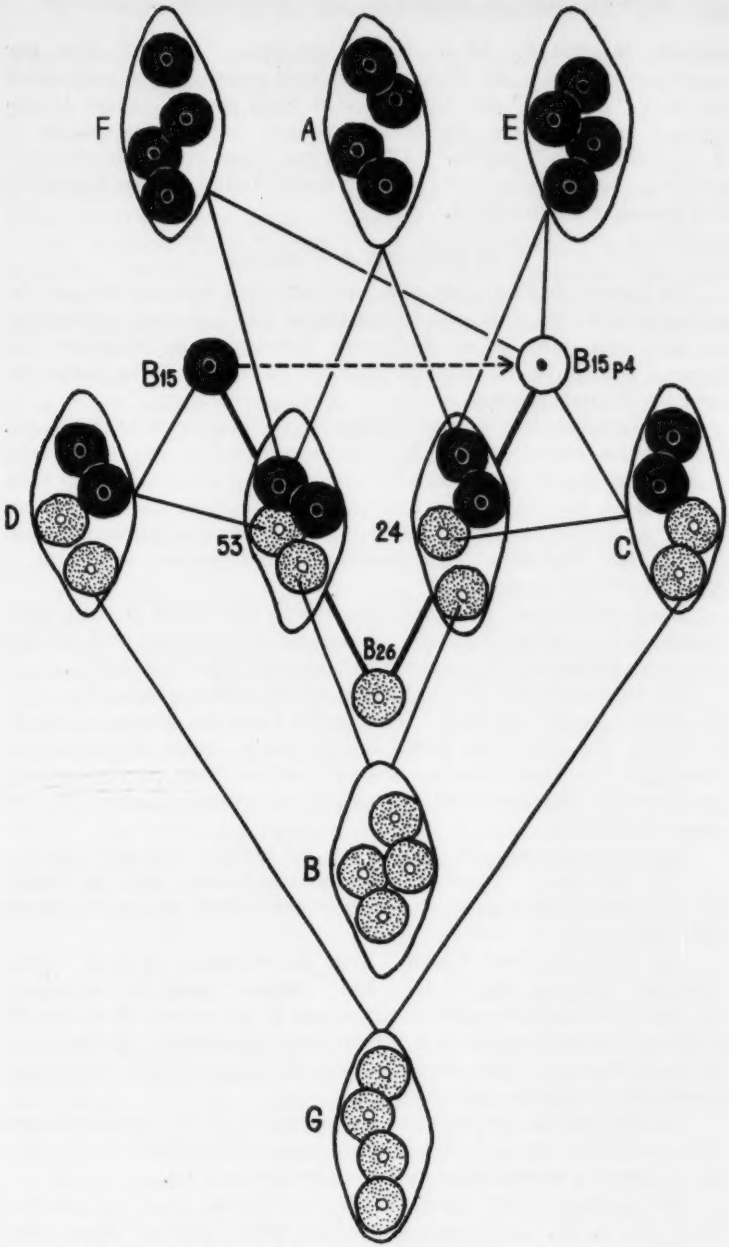


FIG. 1.—Schéma des résultats prévus des croisements. En noir : " grandes " (facteur cytoplasmique actif) ; en grisé : " petites ségrégantes " (facteur cytoplasmique inactif). Noyau noir : gène dominant *R* ; noyau blanc : gène récessif *r*.

seconds, le gène *R*. Le croisement des deux " petites " doit, par conséquent, fournir des diploïdes *r/r* Nadi négatifs et le croisement de deux " grandes " des diploïdes *R/R* Nadi positifs. Etant homozygotes pour *R*, ces diploïdes devraient former des asques à 4 " grandes " : 0 " petites." C'est ce que nous avons effectivement observé. Les croisements 63 et 66 fournissent ainsi à notre hypothèse une première confirmation.

(*d*) *Vérification de l'hypothèse*

La justesse de l'interprétation proposée peut être vérifiée par des croisements variés. Nous en avons réalisé sept qui nous paraissaient les plus importants et en donnerons maintenant les résultats. La figure 1 schématise les résultats *prévus* et permettra de les comparer avec les résultats obtenus.

(i) *Croisement entre " grandes " issues des croisements* 53 *et* 24 (fig. 1, A). Selon notre interprétation, les " grandes " isolées des asques des croisements 53 et 24 portent le même gène dominant *R*. Elles contiennent par ailleurs le facteur cytoplasmique. Le croisement entre " grandes " provenant des deux croisements doit, par conséquent, fournir des diploïdes Nadi positifs et des asques contenant 4 " grandes " : 0 " petites."

Deux croisements (200-201) de ce type ont fourni des diploïdes positifs à la réaction du Nadi. L'analyse de 9 asques a fourni des résultats conformes à la prévision (4 " grandes " : 0 " petites ").

(ii) *Croisement entre " petites " issues des croisements* 53 *et* 24 (fig. 1, B). De même que les " grandes," les " petites issues des deux croisements 53 et 24 doivent, selon notre interprétation, avoir des génotypes identiques (*r*). Le croisement entre ces " petites " doit, par conséquent, produire des diploïdes Nadi négatifs qui ne pourront former que des spores mutantes.

Quinze croisements de ce type ont été réalisés (136-137, 145-152, 160-161, 202-204). Conformément à la prévision, tous ont fourni des diploïdes Nadi négatifs. Aucun de ceux-ci n'a malheureusement sporule.*

(iii) *Backcross d'une " petite " issue du croisement* 24 *à la " petite végétative "* B-15-p4 (fig. 1, C). Les " petites " issues du croisement 24 sont, selon notre hypothèse, identiques à leur parent B-26 dont ils ont reçu à la fois le récessif *r* et le facteur cytoplasmique. Le backcross de ces " petites " à leur parent B-15-p4 doit donc fournir un résultat semblable à celui du croisement 24.

Les résultats de l'expérience (croisements 75 et 77) sont conformes aux prévisions : les diploïdes produits sont Nadi positifs et l'analyse de 18 asques a montré dans tous une ségrégation 2 : 2.

(iv) *Backcross d'une " petite " issue du croisement* 53 *à la " grande "* B-15 (fig. 1, D). La constitution d'une telle " petite " étant, selon

* Ceci est conforme à l'observation que les diploïdes Nadi négatifs ne sporulent jamais (Ephrussi, Hottinguer et Tavlitzki, 1949).

notre interprétation, semblable à celle de la " petite " B-26, ce backcross est strictement équivalent au croisement original (53) et doit fournir un résultat identique.

Conformément à la prévision, le croisement (198-199) a produit des diploïdes Nadi positifs et la ségrégation 2 " grandes " : 2 " petites " a été observée dans 20 asques parmi les 21 analysés. Toutes les 4 spores du 21ème asque étaient " petites."

(v) *Backcross d'une " grande " issue du croisement* 24 *à la " petite végétative "* B-15-p4 (fig. 1, E). Selon notre hypothèse, une telle " grande " tient le gène *R* du parent B-15-p4 et le facteur cytoplasmique de l'autre, la " petite ségrégante " B-26. Le backcross à la " petite végétative " B-15-p4, dont le génotype est également *R*, doit fournir des diploïdes possédant le facteur cytoplasmique actif, donc Nadi positifs. Toutes les spores formées par les asques F_1 contiendront à la fois le gène dominant *R* et le facteur cytoplasmique. Les asques contiendront donc 4 " grandes."

Le croisement (69) a fourni effectivement une culture Nadi positive. Parmi 14 asques analysés, 11 ont donné le résultat prévu ; trois autres ont, au contraire, montré une ségrégation imprévue en 3 " grande " : 1 " petite."

(vi) *Croisement entre " grande " issue du croisement* 53 *et la " petite végétative "* B-15-p4 (fig. 1, F). Une telle " grande " tient le gène dominant *r* du parent " grande " B-15, et ce gène est, par définition, inchangé dans la " petite végétative " B-15-p4 qui ne diffère de B-15 que par la perte du facteur cytoplasmique. Le diploïde hybride doit donc posséder le facteur cytoplasmique et être homozygote pour *R*. Il sera par conséquent Nadi positif et produira des asques à 4 " grandes."

Quatre croisements de ce type ont été réalisés (114, 117-119). Ils ont tous fourni des cultures diploïdes Nadi positives. 27 asques ont été analysés. Les résultats peuvent être résumés comme suit :

Nombre d'asques	Ségrégation
19	4 " grandes " : 0 " petites "
1	3 " : 1 "
2	1 " : 3 "
5	0 " : 4 "

(vii) *Croisement entre " petites " issues des backcross décrits en* (iii) *et* (iv) (fig. 1, G). Les nombreux croisements entre " petites " issues des croisements 53 et 24 n'ayant pas fourni d'asques (voir ii) nous avons essayé de réaliser un croisement analogue (selon l'hypothèse), en utilisant les " petites " issues des backcross décrits en (iii) et (iv).

Douze croisements (399-410) ont été faits dans ce but. Ils ont tous fourni des cultures diploïdes Nadi négatives, mais encore une fois aucune de celles-ci n'a produit d'asques.

(viii) *Etude des spores " aberrantes."* L'ensemble des résultats des expériences décrites ci-dessus est résumé dans le tableau II (colonnes

1 à 5). L'examen de celui-ci permet de constater (*a*) que l'accord entre prévision et observation est toujours parfait quant aux résultats de la réaction du Nadi exécutée sur les cellules diploïdes hybrides ;

TABLEAU 2

Résumé des résultats des croisements

Croisement *	Réaction du Nadi des diploïdes		Ségrégation †		Nature des spores des asques aberrants ‡
	prévue	observée	prévue	observée	
53	+	+	2 : 2	31 (2 : 2)	...
24	+	+	2 : 2	15 (2 : 2)	...
A	+	+	4 : 0	9 (4 : 0)	...
B	−	−	0 : 4	pas d'asques	...
C	+	+	2 : 2	18 (2 : 2)	...
D	+	+	2 : 2	20 (2 : 2)	
				1 (0 : 4)	(pv pv ps pd)
E	+	+	4 : 0	11 (4 : 0)	
				3 (3 : 1)	3 (pv)
F	+	+	4 : 0	19 (4 : 0)	
				1 (3 : 1)	(pv)
				2 (1 : 3)	6 (pv)
				5 (0 : 4)	20 (pv)
G	−	−	0 : 4	pas d'asques	

* Les numéros indiqués ici correspondent à ceux de la figure 1.

† Le premier chiffre donne le nombre de " grandes," le deuxième celui des " petites."

‡ pv "petite végétative" ; ps " petite ségrégante " ; pd " petite double."

(*β*) qu'à côté des asques du type prévu, d'autres ont été trouvés, dans trois des sept croisements, qui montrent des ségrégations imprévues et que nous allons qualifier " d'aberrants."

Etant donné que tous les asques aberrants contiennent un *excés de spores mutantes*, l'hypothèse la plus simple, susceptible de rendre compte, dans le cadre de l'interprétation adoptée, de leur apparition, consiste à admettre qu'elles sont dues à la perte du facteur cytoplasmique postulé. Les " petites " spores surnuméraires, quoique apparues à la suite de la méïose, seraient essentiellement ce que nous avons appelé des " petites végétatives." Elles contiendraient donc toutes le gène dominant *R*.

La vérification de cette hypothèse a été entreprise de la façon suivante. Le clone dont on cherche à déterminer la nature est croisé, par la méthode de Lindegren : (*a*) avec un clone standard de " petites végétatives " ; (*b*) avec un clone standard de " petites ségrégantes."* Les cellules diploïdes formées sont soumises au test du Nadi.

Si le croisement (*a*) donne un test positif et (*b*) un test négatif, le clone examiné est une " petite ségrégante " (*r*), puisque la " petite

* Nous avons pris comme clones standard de " petites végétatives " de signe opposé, les souches 1-A_2 et B-15-p4 mentionnées plus haut. Pour standards de " petites ségrégantes " nous avons choisi deux " petites ségrégantes " (*19-a* et *19-c*) d'un meme asque du croisement 53.

végétative " avec laquelle il a été croisé était dépourvue de facteur cytoplasmique. Il a donc dû apporter le facteur cytoplasmique. Si, au contraire, le croisement (*a*) est Nadi négatif et le croisement (*b*) Nadi positif, le clone examiné est classé comme " petite végétative " puisqu'il a dû apporter le gène dominant *R* qui manquait à la " petite ségrégante " avec laquelle il a été croisé.

Notons incidemment qu'une troisième possibilité existe également : une levure pourrait à la fois être dépourvue de facteur cytoplasmique et porter le gène récessif *r*. Dans ce cas, les deux croisements (*a*) et (*b*) donneraient des résultats négatifs. Nous appellerons ce type de mutant " petite double."

La méthode que nous venons d'indiquer a été appliquée à tous les clones mutants provenant des asques aberrants. Les résultats du test sont donnés par la dernière colonne du tableau 2. On voit que toutes les spores exceptionnelles résultent de la perte du facteur cytoplasmique et que la ségrégation génique dans tous les asques aberrants est conforme à la prévision.

4. DISCUSSION

Comme il a été dit dans l'Introduction, le présent travail a été entrepris avec l'espoir de mettre en évidence une interaction entre le génome et le facteur cytoplasmique dont la présence dans les cellules de la levure est indispensable à la synthèse de certains ferments respiratoires.

Cet espoir n'a pas été déçu.

(*a*) Notre travail a tout d'abord apporté une nouvelle preuve de l'existence de ce facteur cytoplasmique. Les résultats permettent en effet d'affirmer que la souche " petite végétative " B-15-p4 ne diffère de la souche-mère " grande " B-15, que par l'absence d'un facteur cytoplasmique autoreproductible. Nous retrouvons donc, entre ces deux souches de la levure " Boulangerie II," des relations exactement semblables à celles qui ont été mises en évidence entre deux souches de " Yeast Foam " (Ephrussi, Hottinguer et Tavlitzki, 1949).

(*b*) Dans le travail ci-dessus, nous avons pu pousser l'analyse plus loin. Les croisements de la " petite ségrégante " B-26 avec la " petite végétative " B-15-p4 et avec la " grande " B-15 ont fourni des résultats qui peuvent être expliqués si l'on admet : (α) que les " petites ségrégantes " diffèrent des " grandes " par un seul gène dont elles contiennent l'allélomorphe récessif ; (β) qu'elles diffèrent des " petites végétatives " à la fois par ce même gène et par le facteur cytoplasmique qu'elles possèdent (comme les " grandes ") et dont les " petites végétatives " sont dépouvures. Une série de croisements entrepris pour vérifier ces hypothèses ont montré que les génotypes observés sont toujours en accord avec ceux postulés ; et que les quelques aberrations phénotypiques rencontrées s'expliquent raisonablement par la perte du facteur cytoplasmique.

Tout se passe donc comme si la synthèse des ferments respiratoires dépendait de la présence simultanée d'un facteur cytoplasmique autoreproductible et d'un gène dominant, en absence duquel le facteur cytoplasmique demeure inactif.

Le facteur cytoplasmique de la levure serait donc " autonome " dans sa reproduction ; il dépendrait au contraire du génome dans sa fonction. Ces propriétés le rapprochent singulièrement des chloroplastes des plantes.*

(*c*) Dans le présent travail, nous avons, pour la première fois, introduit une distinction motivée entre " petites végétatives " et " petites ségrégantes." La " petite " 1-A_2 de Yeast Foam, étudiée dans le travail cité ci-dessus, appartient de toute évidence à la première de ces catégories. Cela explique la disparition du caractère mutant dans les croisements avec une " grande " et dans les quatre backcross effectués par les auteurs ; et sa disparition dans le croisement (1) décrit dans le paragraphe III (*a*). Tout récemment, nous avons réussi à croiser une " petite ségrégante " (No 53-19, provenant donc du croisement 53) de B-II avec la souche 1-A_2, " petite végétative " de Y.F. Ce croisement est suivi d'une ségrégation régulière 2 " grandes " : 2 " petites," ce qui montre que la souche 1-A_2 de Y.F. contient le même gène dominant *R* que la souche B-15 de B-II. Il est donc évident que le facteur cytoplasmique est, dans les deux souches, soumis à un contrôle génique simple. Ces faits rendent probable la suggestion faite dans l'Introduction, selon laquelle la fréquence plus haute de spores mutantes dans les asques de B-II serait due à ce que cette souche est hétérozygote pour un gène récessif.

(*d*) Dans ce qui précède, nous avons, à dessein, utilisé le terme " facteur cytoplasmique " qui ne préjuge pas de la nature continue ou discontinue de ce constituant. On se rappelle que l'hypothèse d'un facteur héréditaire extranucléaire chez les levures, introduite par Ephrussi et ses collaborateurs, attribue à ce facteur une structure particulaire (voir Introduction).

Notre travail apporte-t-il des éléments nouveaux à ce problème de la structure du facteur cytoplasmique ? Il nous paraît que deux constatations faites au cours des recherches relatées ici plaident en faveur de la nature discontinue, c'est-à-dire particulaire, du facteur cytoplasmique.

C'est d'abord la fréquence élevée des ségrégations " aberrantes," qui s'expliquent, comme nous avons pu le montrer, par la perte du facteur cytoplasmique. Considérons en particulier les asques des types 3 : 1 et 1 : 3. On ne voit pas comment un facteur cytoplasmique continu, c'est-à-dire imprégnant d'une façon homogène le cytoplasme de la cellule-mère de l'asque, pourrait se répartir inégalement au cours de la sporogénèse.

* L'interprétation proposée est celle qui nous paraît aujourd'hui la plus vraisemblable. Nous tenons cependant à souligner qu'une autre interprétation de nos résultats expérimentaux est possible si l'on admet : (1) la dépendance de la reproduction des particules cytoplasmiques du gène *R* ; (2) un effet retardé de ce gène ; et (3) un seuil de concentration des particules dans leur action physiologique.

C'est ensuite, la similarité entre les propriétés de notre facteur cytoplasmique et celles des plastes des plantes vertes.

(*e*) Le mode d'origine des asques " aberrants " sera discuté dans un prochain mémoire.

5. RÉSUMÉ

1. Dans la souche " Boulangerie II " de *S. cerevisiæ* des mutants à déficience respiratoire (" petites ") apparaissent souvent, d'une part au cours de la reproduction végétative (" petites végétatives "), et d'autre part à la suite de la méïose (" petites ségrégantes "). La fréquence des spores mutantes est d'environ 44 pour cent. Ce pourcentage est suffisamment proche de 1/2 pour qu'il soit légitime de se demander si la souche n'est pas hétérozygote pour un couple de gènes et si le caractère mutant des " petites ségrégantes " n'est pas sous le contrôle d'un gène mendélien.

2. L'étude des deux croisements possibles entre une levure normale (" grande " B-15), une " petite végétative " (B-15-p4) qui en dérive, et une " petite ségrégante " (B-26) a fourni les résultats suivants :

(*a*) Le croisement de la " grande " B-15 avec la " petite ségrégante " conduit à la formation de cellules diploïdes normales ; les asques formés par celles-ci contiennent 2 " grandes " et 2 " petites " spores.

Chez les " petites ségrégantes," le caractère " petite colonie " est donc dû à un gène mendélien récessif.

(*b*) Le croisement de la " petite végétative " B-15-p4 avec la " petite ségrégante " B-26 produit des cellules diploïdes normales, et les asques formés par celles-ci contiennent encore une fois 2 " grandes " et 2 " petites " spores.

Dans ce croisement, on obtient, par conséquent, la reconstitution d'une cellule normale par la fusion de deux cellules mutantes. Ceci prouve tout d'abord que des phénotypes identiques peuvent cacher des idiotypes différents ; cela montre ensuite que les deux souches croisées doivent différer par quelque chose d'autre que la paire de gènes dont on observe la ségrégation dans les asques F_1.

3. Ces résultats nous ont conduit à proposer l'interprétation suivante : les " petites végétatives " diffèrent des " grandes " par la perte d'un facteur cytoplasmique nécessaire à la synthèse des ferments respiratoires ; les " petites ségrégantes " en diffèrent par un gène récessif en présence duquel le facteur cytoplasmique est inactif.

4. Afin de vérifier cette hypothèse et, en particulier, son corrollaire selon lequel les génotypes des " petites végétatives " et des " grandes " sont identiques, sept types de croisements différents ont été entrepris. L'analyse des asques résultant de ces croisements fournit des génotypes conformes à la prévision. Des phénotypes exceptionnels ont cependant été observés, dont l'étude conduit encore une fois à postuler l'intervention d'un facteur cytoplasmique autoreproductible.

5. Au total, le présent travail conduit à la conclusion que la synthèse de certains ferments respiratoires requiert la présence simultanée d'un facteur cytoplasmique autoreproductible et d'un gène dominant.

6. SUMMARY

1. In the strain " Boulangerie II " of *Saccharomyces cerevisiæ* mutants often appear which lack certain respiratory enzymes (" little colony " or " little "). They arise in the course of vegetative reproduction (" vegetative littles ") as well as following meiosis (" segregational littles "). The frequency of the mutant spores is about 44 per cent., *i.e.* so close to 1/2 that the question was raised as to whether the strain is heterozygous for a gene pair, and the mutant character controlled by a mendelian gene.

2. A study of the two crosses possible between a normal clone (" big," B-15), a " vegetative little " (B-15-p4) derived from it, and a " segregational little " (B-26), has given the following results :

(*a*) The cross of " big " B-15 with the " segregational little " B-26 gives rise to normal diploid cells which form asci containing 2 " big " and 2 " little " spores.

Consequently, the character " little " is due to a single mendelian recessive gene in the " segregational littles."

(*b*) The cross between the " vegetative little " B-15-p4 and the " segregational little " B-26 also gives normal diploid cells, the asci from which likewise contain 2 " big " and 2 " little " spores.

Thus, in this cross, the fusion of two mutant cells leads to the reconstitution of a normal cell. First of all this shows that the two phenotypically identical cells used in the cross conceal different idiotypes. Secondly it demonstrates that the two strains differ from each other by something other than the gene pair whose segregation takes place in the F_1 asci.

3. These results have led to the following interpretation : the " vegetative littles " differ from the " bigs " or normals by the loss of a cytoplasmic factor which is necessary for the synthesis of certain respiratory enzymes ; the " segregational littles " differ from normals by a single recessive gene in the presence of which the cytoplasmic factor is inactive.

4. Seven different kinds of crosses were undertaken in order to test this hypothesis, and in particular its corollary, according to which the " vegetative littles " and the " bigs " have the same genotypes. The genotypes of the segregants in the asci of these crosses were found to be those predicted by the above hypothesis. However, some exceptional phenotypes were observed, study of which has again led to the postulation of a self-reproducing cytoplasmic factor.

5. As a whole, the present work leads to the conclusion that the synthesis of certain respiratory enzymes in yeast requires the simultaneous presence of a self-reproducing cytoplasmic factor and a dominant gene.

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IS GENE DISPERSION NORMAL ?

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INTRODUCTION

In the early theoretical studies of population genetics (Wright, 1931) the simplest cases naturally were considered. Populations were taken to be internally and externally panmictic. That is mating was completely at random within each population and finite migration occurred at random between otherwise isolated populations. Such is, of course, an ideal situation never realised in natural populations, though examples of true *internal* panmixis exist in crop plants in which all the seed from a population is thoroughly mixed before sowing the next generation, and in animals such as fish in ponds, where the radius of the occupied space is much smaller than the mean distance travelled per generation.

Apart from such exceptions there will always be a correlation within populations between the proximity of individuals to each other and their genetic relationship. There will be a similar correlation between the proximity of populations to each other and their genetic relationship. And since the greater the proximity the greater the chances of mating, there will always be a certain amount of inbreeding relative to the population as a whole or the group of populations, whatever may be the size and number of the populations.

In later years (Wright, 1943, 1946) allowance was made for this by considering the structure of continuous populations. The symbol N, originally the size of a panmictic population, was carried forward into the new situation in a new role. It now represented the sizes of sub-populations into which a continuous population was theoretically divisible. In order to avoid implying any particular breeding behaviour these sub-populations were referred to as " neighbourhood units." The magnitude of the new N was such as to give, with random dispersion within the neighbourhood, an average gene dispersion equivalent to that obtaining in the non-panmictic continuous population.

It may be noted in passing that fixation and drift are much less dependent on the breeding behaviour itself than on the gene dispersion. Hence the introduction of the neighbourhood concept to cover everything from true panmixis to populations of self-fertilising plants where the genes are always dispersed as zygotes.

N is given by the number of individuals within a circle of radius 2σ where σ^2 is the variance of gene dispersion within the species. Malécot

(1948), dealing with the same problem, relates the variation within a continuous population directly to σ^2 without the introduction of the neighbourhood concept. From the key position that gene dispersion holds, its estimation under natural conditions for various organisms is clearly of importance to population genetics. Such an estimation has been made for *Drosophila pseudoobscura* by Dobzhansky and Wright (1943 and 1947).

There are, however, other important parameters of continuous populations. Such are the mean degree of inbreeding and the frequency distribution of degrees of inbreeding. In diœcious organisms and self-incompatible plants the mean degree of inbreeding is adequately given by 1/N, N being calculated from the gene dispersion. In organisms with varying degrees of self-fertilisation this is not so. In the extreme case of complete self-fertilisation, inbreeding is unity whatever the value of N. Under no circumstances can the frequency distribution of degrees of inbreeding be expressed merely in terms of 1/N. For this we must know not only the degree of dispersion (the variance) but the nature of the dispersion ; whether it is skew or has abnormal kurtosis (the fourth degree statistic).

In the absence of evidence to the contrary it is natural to assume that gene dispersion should follow the normal distribution curve. This assumption has been made, for example, by Wright (1943, 1946) and by Haldane (1948) though in no instance was normality fundamental to the argument. It is desirable, however, whenever the data make it possible, to test the nature of gene dispersion. Skewness may be expected to be frequent. It will arise whenever there is a tendency for migration in one direction ; say downwind.

The standard method for the estimation of kurtosis involves fourth degree statistics. If k_2 is the second moment (variance) and k_4 the fourth moment, the kurtosis of a distribution is given by the expression k_4/k_2^2-3. For a normal distribution $k_4/k_2^2 = 3$ and kurtosis is zero. With a platykurtic distribution in which the " shoulders " of the frequency curve have increased at the expense of the peak and " tails," $k_4/k_2^2<3$ and kurtosis is negative. With a leptokurtic distribution peak and tails are exaggerated, $k_4/k_2^2>3$ and kurtosis is positive. The three kinds of kurtosis are illustrated in fig. 1. Though this method of estimation of kurtosis is mathematically most directly related to the properties of normal distributions, a practical difficulty involved in its application is that k_4 is extremely sensitive to incompleteness of sampling area. Values farthest from the mean make disproportionately large contributions to k_4 and so with a finite sampling area there is always a possibility that there may be a loss of extreme values (a truncation of the tails of the distribution), a reduction in the ratio k_4/k_2^2 and a negative bias in the estimation of kurtosis. This is not merely a theoretical possibility. Dobzhansky and Wright (1947) found that high positive kurtosis on the first day, when the sampling area was most likely to extend beyond the limits

of the distribution of the flies, steadily diminished with succeeding days, when it became almost certain that the limits of distribution exceeded the sampling area. In some cases the original positive kurtosis was thereby transformed into a negative one. An alternative, if empirical, method of studying kurtosis has now been developed (Bateman, 1947). It is limited in application to the detection of marked kurtosis, but it is not dependent on sampling covering the complete distribution. This is the method of regression.

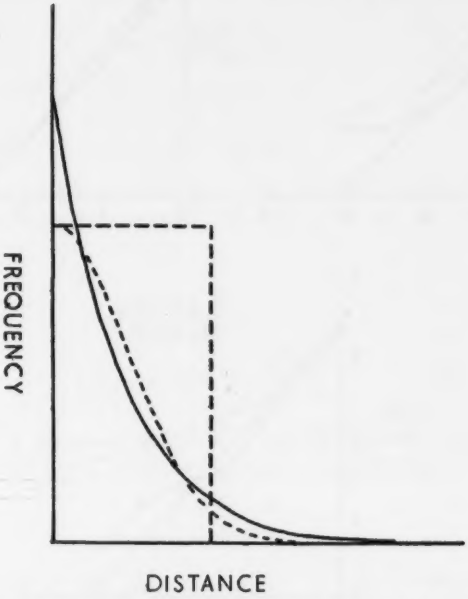

FIG. 1.—Half the curve for normal dispersion in one dimension is represented by the curved broken line. The Y axis therefore marks the centre of dispersion. The entire curve has positive kurtosis (leptokurtic). It shows abnormally high values for both short and long extreme distances. The rectangle represents an extreme form of negative kurtosis (platykurtic). It also represents Wright's neighbourhood unit, used as a model for gene dispersal in a continuous population. The length of its base is 2σ, its height is the density of the population and its area is $\frac{1}{2}$N.

The normal distribution can be expressed empirically as $y = ge^{-bx^2}$, where x is the deviation from the mean and g and b are constants. Logarithmically this becomes $\log y = a - bx^2$. Any normal distribution can therefore be adequately represented by the regression of $\log y$ on x^2. Kurtosis will modify the regression line by increasing or decreasing the curvature (see fig. 2). Leptokurtosis will straighten the curve and even, if extreme, produce a concave instead of a convex curve. Such a change of curvature is equivalent to a change in the power of x from 2. If the power were 1 the curve would be straight,

if $\frac{1}{2}$, concave. If a distribution is leptokurtic the best fit for the regression of $\log y$ on x^n will be given when n is significantly less than 2.

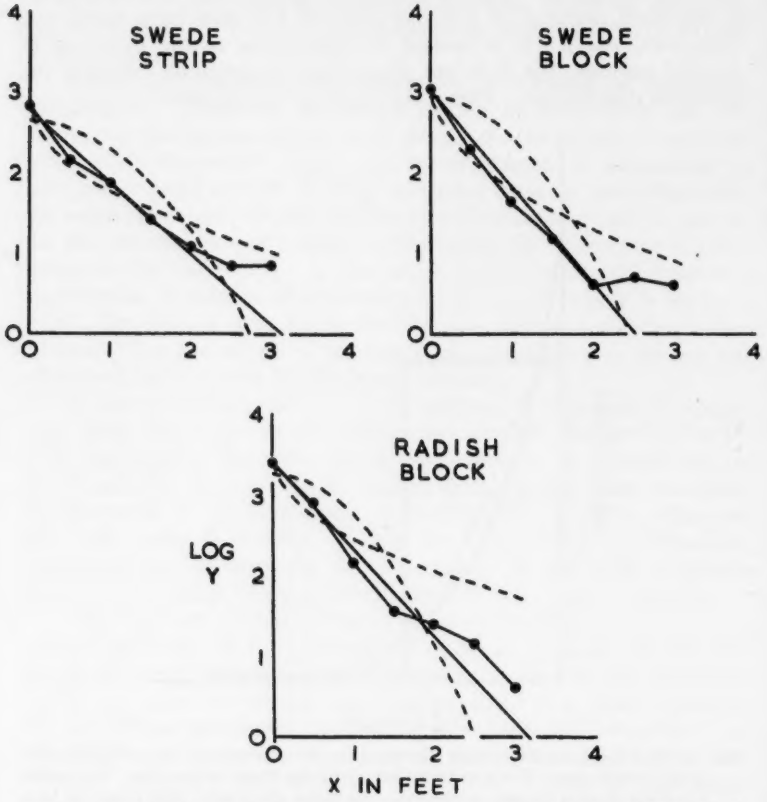

FIG. 2.—Semilogarithmic curves are given for the frequency distribution of flight lengths of pollinating insects on two crops. This corresponds to the half of a distribution curve shown in fig. 1. The observed frequencies are shown as polygons. Also shown for comparison are the regression lines for log Y on X^2 (convex broken line) X (straight entire line) and $\sqrt{X}$ (concave broken line). The ideal curve would appear to be somewhere between the X and $\sqrt{X}$ regressions.

DISPERSAL OF ANIMALS

Examples of this method applied to the flight of pollinating insects have already been given (ibid.). The regressions of $\log y$ on x and $x^{\frac{1}{2}}$ were definitely superior to those on x^2. The $x^{\frac{1}{2}}$ regression was slightly but not significantly better than the x regression. Further data have now been obtained and can be analysed in the same way. Some refinements have been introduced. Firstly, only the projection of

each flight on to a predetermined line was measured. Thus the data were immediately in the form of one-dimensional distribution and did not require conversion from a two-dimensional distribution as previously. Secondly, flights longer than three feet have been excluded from the regression since it was felt that errors of observation rapidly increase beyond this distance. Thirdly, to counteract the high error variance of $\log y$ when y is small (rising to infinity when $y = 0$) a system of weighting was introduced. The information (invariance) about $\log y$ is related to the information about y in the following way :—

$$I_{\log y} = \frac{dy}{d \log y} I_y$$

$$\frac{dy}{d \log y} = y \log_e 10$$

Therefore $$I_{\log y} = y^2 \log_e{}^2 10 \, I_y$$

V_{y1} is given by the usual formula for binomial distributions adapted to the present circumstances ($p \equiv y_1$, $q \equiv (\Sigma y - y_1)$ and $n \equiv \Sigma y$) *i.e.* $\frac{y_1(\Sigma y - y_1)}{\Sigma y}$

Hence $$Iy_1 = \frac{\Sigma y}{y_1(\Sigma y - y_1)}$$

and $$I_{\log y_1} = \log_e{}^2 10 \, \frac{\Sigma y \, . y_1}{\Sigma y - y_1}$$

$\text{Log}_e{}^2 10$ is a common factor and can be ignored when weighting. Each observed $\log y_1$ is then weighted by multiplying by

$$\frac{\Sigma y \, . y_1}{\Sigma y - y_1}$$

Fig. 2 shows the observed values of $\log y$ compared with the regression lines corresponding to the weighted regressions on x^2, x and $x^{\frac{1}{2}}$. The goodness of fit of these regressions can be judged from table 1 by the comparison of their remainder sums of squares. The inadequacy of the x^2 regression (corresponding to a normal distribution) is clearly demonstrated, both graphically and statistically. There seems little to choose between the x and $x^{\frac{1}{2}}$ regressions. In fact the ideal regression would appear to be one in which the index of x was about $\frac{3}{4}$. This is stronger evidence than hitherto presented of the leptokurtic nature of the dispersion of foraging bees.

An independent set of data obtained by quite different methods is furnished by Dobzhansky and Wright (1947) on the dispersal of mutant flies of *Drosophila pseudoobscura* from their point of liberation. Semilogarithmic curves for some of the days are given in fig. 3. The flies were caught in traps placed in a straight line at intervals of 20 metres. Kurtosis as measured by $k_4/k_2{}^2$ was strongly positive on

TABLE 1

A comparison of the goodness of fit obtained by the regressions of log y *on* x^2, x *and* $x^{\frac{1}{2}}$, *where* x *is the flight length of hive bees and* y *is their frequency.*

Data from	Regression on	Regression sum of squares	Remainder sum of squares	Degrees of freedom	Variance Ratio P
Swede strip	x^2	129·6305	82·9529	5	22·50<0·01
	x	201·2890	11·2944		3·06<0·2
	$x^{\frac{1}{2}}$	208·8961	3·6873		
Swede block	x^2	162·9438	136·3764	5	13·41<0·01
	x	285·7909	13·5294		1·33 high
	$x^{\frac{1}{2}}$	289·1509	10·1693		
Radish block	x^2	450·6367	231·7158	5	19·40<0·01
	x	670·4069	11·9456		
	$x^{\frac{1}{2}}$	597·6855	84·6670		7·09<0·05

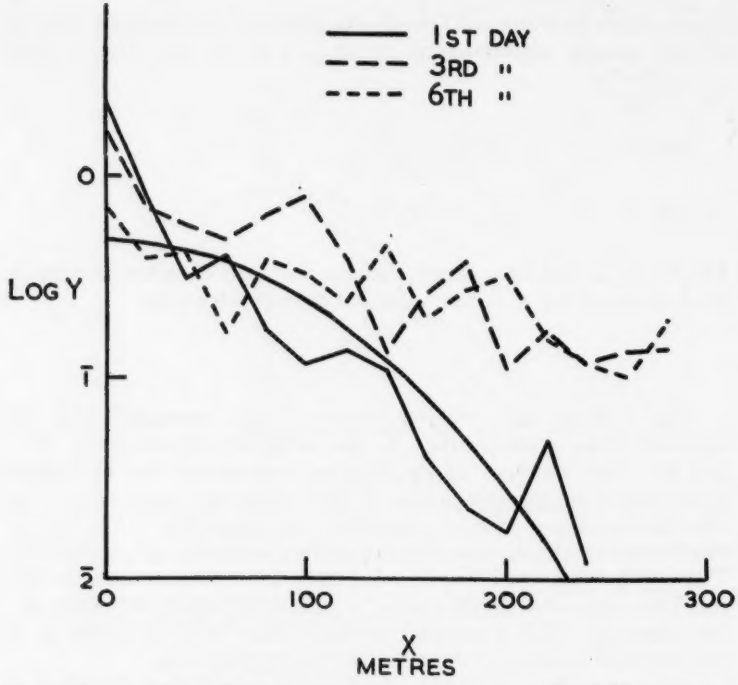

FIG. 3.—Semilogarithmic curves for the distribution of mutant flies one, three and six days after liberation at distance 0 (Dobzhansky and Wright's data). Also shown (continuous curve) is the expected first day's distribution on the basis of normal distribution. It gives expectations which are too low at the point of liberation and over 200 metres, but too high for intermediate distances. Casual inspection indicates that good fits for the data on all three dates would be obtained by a linear regression of log. frequency on distance.

the first day and diminished steadily to the sixth day. The main difference between this fly data and the above bee data is that while the latter were based on single flights, the shortest interval in the former was a whole day during which there would be numerous flights per fly. The positive kurtosis of the flies is therefore even more unexpected. The authors recognised that the diminution of kurtosis with succeeding days might be due to increasing incompleteness of sampling (discussed above). In a private communication, however, Wright has held that the main factor reducing kurtosis was the normalising effect of subsequent flights. He assumes that consecutive flights of *Drosophila* are uncorrelated. If that were so it would be very surprising for the distribution of flies after the first whole day's flights still to be strongly leptokurtic in spite of the normalising effect to be expected from lack of correlation between the many successive flights during that day.

The regression method has been used on the above data to see whether any further light could be thrown on the question. The frequencies were again weighted before calculating the regression, this time by dividing every frequency of mutant flies by the total number of wild type flies (the original population) at that site. This allowed for the very noticeable variation in attractiveness of the sites and gave much smoother frequency curves. Only traps near to the point of liberation gave high enough counts of mutant flies to be used in the regression method.

The results are given in table 2. The superiority of the x regression over x^2 again demonstrates the leptokurtosis. This in spite of the

TABLE 2

Day	Regression on	Regression sum of squares	Remainder sum of squares	Degrees of freedom	Variance ratio	P
1	x^2	3·682968	1·359011	9	2·40	0·1
	x	4·476328	0·565651			
2	x^2	3·650894	1·482418	14	1·85	0·1
	x	4·334005	0·799307			
3	x^2	2·673754	0·971419	14	1·57	0·2
	x	3·026842	0·618331			
4	x^2	3·211919	0·575768	14	1·35	0·3
	x	3·359720	0·427967			
5	x^2	2·424195	0·815767	14	1·38	0·3
	x	2·647765	0·592197			
6	x^2	1·248294	0·588813	14	1·11	0·7
	x	1·307332	0·529775			

restriction of sampling to the proximal zone. The leptokurtosis cannot therefore be due merely to long " tails " to the distribution curve, but expresses itself also in other parts of the curve. Unfortunately no

single day's observations gives a significantly better x than x^2 regression. If, however, the probabilities for each day are multiplied a compound probability of 0·0001 is obtained. Such a compounding is not strictly justifiable unless the observations are independent of one another, which these are not, being based on a constant fly population. The very low compound probability does, however, give some confidence that the leptokurtosis is real.

The regression appears to agree with the fourth moment in showing a steady approach to normality with successive days. That is, the ratio of the remainder sums of squares for the x^2 and x regressions becomes steadily less significant. There is another purely statistical explanation for this. With each succeeding day the slope of the distribution diminishes. The sum of squares which is available for any linear regression will therefore diminish, while the true error sum of squares will presumably remain about constant. The remainder sum of squares will therefore increase relative to the amount taken out by any regression and to the differences between the amounts taken out by any two regressions. Correspondingly it will be increasingly unlikely that any one linear regression should become significantly superior to any other.

In conclusion, the application of the regression method to the *Drosophila* observations, although confined only to the proximal part of the distribution, confirms the leptokurtosis originally found by an unrelated method. It also appears to confirm a day-to-day reduction in kurtosis, though other interpretations of the results from both methods of analysis are possible.

WIND DISPERSAL

Semilogarithmic curves for pollen dispersal (Bateman, 1947) show straight or even slightly concave curves relating $\log y$ to x (therein called $\log P$ and D respectively). They are also fitted well by regression lines using powers of x equal to or less than unity. In view of the general similarity of all wind dispersal it is probable that spores would be distributed in a similarly leptokurtic way.

CROSS-POLLINATION

Cross-pollination is here used in the sense of pollination between two populations or groups of plants which are separated by distance x.

The distribution of cross-pollination in wind-pollinating species will be merely an extension of pollen dispersal and the same leptokurtosis will apply. One limitation which will rarely have an effect is that there is a maximum degree of cross-pollination *viz.* 100 per cent. This would at most cause a slight depression of the peak of the distribution, without affecting the tails.

The situation in insect-pollinated species is more complex. In the first place it is dependent on the flight of the pollinating insects

which, as we have seen, is leptokurtic. But it can be shown (ibid.) that as with each visit the insect not only deposits pollen but replaces part of its load by pollen from the flower it is visiting, the leptokurtosis tends to increase still further. Thus if the distribution of the insect were such as to give a straight line relation between $\log y$ and x, the curve for cross-pollination at distance x from the source would be concave. This expectation was confirmed by the data obtained from experiments on cross-pollination.

THE ORIGIN OF LEPTOKURTOSIS

The following types of gene dispersion have thus been shown to be leptokurtic : automatic movement of airborne insects (Dobzhansky and Wright, 1943, 1947 ; Bateman, 1947 and present paper) ; water fleas (Brownlee, 1911) ; passive movement of airborne organisms (Gregory, 1945 ; Bateman, 1947) ; dispersal of pollen by insects (Bateman, 1947). There remain a number of important types of gene dispersion for which the necessary information is not yet available : the movement of terrestrial animals and birds ; the movement of plankton ; and the dispersal of seeds by explosive and other automatic mechanisms. When such a varied collection of known dispersal types has been shown to be consistently leptokurtic there is no reason to doubt that the balance would be similarly non-normal. It is possible, therefore, that gene dispersion is as a general rule, leptokurtic.

There is an apparent contradiction between such a situation and the general theory of normal distributions. Under this theory normality should result whenever a large number of uncorrelated influences are at work. The contradiction forces us to the conclusion that neither the passive nor active movement of organisms is at random. The leptokurtosis observed in the distribution of pollinating insects after single flights suggests a tendency for flights to be either short or long, though the smoothness of the curve shows that there are also all intermediate flight lengths. A possible explanation would be that some flights involved the continuation of an extremely localised forage whereas others were exploratory and ranged over a wider field.

Even if the distribution of *Drosophila* after one flight had been similarly leptokurtic, only a strong correlation between consecutive flights in direction, length, or both would maintain the leptokurtosis after one day's flying. Since the leptokurtosis remained in evidence for several days the correlation between flights must have remained over that period. Unless, however, the correlation were absolute there would be a day-by-day drift to a normal distribution. If we were to classify the individual flies according to their flight lengths into sedentary and wandering classes the classification would tend to become steadily more blurred with extension of time. A closely parallel classification of hive bees, varying with time, has been postulated by Butler (1945). The leptokurtosis of pollen dispersion

by insects is merely an accentuation of that for the insects themselves and requires no separate consideration.

The explanation of the leptokurtosis produced by wind dispersal is to be found in a theoretical discussion of the subject in which the problem is treated as an example in aerodynamics (Gregory, 1945) and is discussed in terms of atmospheric turbulence or eddy diffusion. Gregory's general formula has been expressed in simpler terms (Bateman, 1947) as

$$y = \frac{ge^{-\left[\frac{cx^{(1-\frac{1}{2}m)}}{1-\frac{1}{2}m}\right]}}{x^{(1+\frac{1}{2}m)}}$$

Where y is the number of particles deposited per unit area, x is the distance from the source, g and e are empirical constants and m is the turbulence factor, varying between the limits of 1·24 and 1·76. The power of x to which e is raised, $1-\frac{1}{2}m$, therefore varies between the limits of 0·38 and 0·12 respectively compared with 2 in a normal distribution. The leptokurtosis produced by the lower power of x in the numerator is still further enhanced by the presence of x in the denominator. All gene dispersion depending on wind could therefore be expected to be highly leptokurtic. Such a dispersion would be characteristic of fungal spores, passively borne insects, pollen grains and wind-borne seeds, amounting in all to a large cross-section of all forms of gene dispersal.

It remains to be seen what genetic effects, if any, leptokurtic gene distribution would have. Such effects would probably arise out of the fact that, compared with a normal distribution having the same standard deviation (same overall degree of inbreeding), leptokurtosis produces more breeding between close relatives and simultaneously more breeding between distant relatives.

It is also possible that the " migration " considered by Wright as a category distinct from dispersal within the neighbourhood unit would prove to be adequately expressed as the tails of the leptokurtic distribution and therefore a part of the same gene dispersal system.

SUMMARY

A number of different forms of gene dispersal : the foraging of bees ; the flight of *Drosophila* over several days ; the dispersal of spores or pollen by wind ; cross-pollination by bees, are all shown to give leptokurtic distributions. That is, they differ from normal distributions in having higher proportions of short and long range, as compared with medium dispersal. The implication is that many if not most methods of gene dispersal produce such distributions.

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THE GENETICS OF TRISTYLY IN *OXALIS VALDIVIENSIS*

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THE efficiency of tristyly as a crossbreeding system depends, as Fisher and Mather (1943) pointed out, on the equilibrium of the three forms in a population. The inheritance of these forms is, therefore, of considerable interest. In *Lythrum salicaria*, Fisher and Mather (1943) disproved East's theory of linked lethals, and demonstrated tetrasomic inheritance. The purpose of this paper is to report the differences and similarities found, during the course of work carried out at Cambridge since 1946, in *Oxalis valdiviensis*, a wild species from Chile.

The three forms, which have been described by Darwin (1880) and Barlow (1913), differ in respect of the length of the style, the length of the two whorls of stamens, and the size of the pollen grains. Each form has two different lengths of stamens and a style of yet a third length, so that the anthers of the long-styled flowers are at the mid and short level, of the mid-styled form, at the long and short level, and, of the short-styled form at the long and mid level. For legitimate pollination it is essential that pollen from anthers of the same level as the style be used, each form can, therefore, be legitimately fertilised by both the other forms.

With both these species, Darwin showed that when illegitimate pollinations were made, *i.e.* using pollen from a different level to that of the style, which includes selfing, very few, if any, capsules resulted. With *Oxalis valdiviensis*, he quotes Hildebrand (1871), who obtained 6 capsules from 255 pollinations with an average of 3·83 seeds. On the other hand, from 138 legitimate pollinations, he obtained 138 capsules with an average of 11·33 seeds in each.

The facts established by C. Darwin (1880) and Barlow (1913 and 1923) were that the long-styled form is the recessive, since only long-styled plants were produced from illegitimate self-fertilisations and cross-fertilisations between two Longs ; that two loci are involved —that for short style being epistatic to that for mid, so that short-styled plants can carry the mid allele, showing this in their progeny, while mid-styled plants cannot carry the short gene.

This two-factor hypothesis, though first published by von Ubisch (1921), was, in fact, the hypothesis on which Barlow had previously based her work published in 1923 (see East, 1927). East's investigations on *Lythrum* led him to write, " One can hardly examine this

evidence without feeling that the two-factor hypothesis is an approach to the truth. Yet it is not the whole truth." He then went on to postulate that " Mid is conditioned by duplicate linked factors, M_a and M_b that are lethal in the homozygous condition." In effect a three-factor hypothesis. It was only when Fisher and Mather (1940) showed the non-lethality of the mid factor, and, in 1942, polysomic inheritance, that the two-factor hypothesis, in its new form, was firmly established in respect of *Lythrum*.

Throughout our experiments we adopted the open pollination method used by Fisher and Mather (1943) in their experiments with *L. salicaria*. This consists of surrounding the plants to be fertilised by plenty of suitable legitimate pollen plants in an isolated plot, and allowing insects to do the pollinations. The proportion of stray pollen may be verified from the progenies, and in *Lythrum* was found to be only 1 per cent. of that available (Fisher and Mather, 1943). It is interesting also that illegitimate unions occurring with appreciable frequency may be easily detected. An abundant supply of seed with the opportunity of growing sufficiently large progenies is the outstanding practical advantage.

DISOMIC INHERITANCE

To find out whether inheritance is disomic or polysomic the procedure of Fisher and Mather (1943) in establishing tetrasomic inheritance in *L. salicaria* was followed.

Seed was obtained from the Botanic Garden in Cambridge in 1946. The resulting plants were classified for style length and twelve Shorts set out in an isolated plot and surrounded by sixteen long-styled plants as pollen parents. Seed was harvested from the Shorts and 12 progenies of 120 each grown the following summer. These were scored for style-length (table 1).

TABLE 1

Progenies grown in 1947 testing 12 Shorts

Plant Number	L	M	S	Total	Plant Number	L	M	S	Total
S46.A	52	...	65	117	S46.B	8	49	62	119
S46.G	66	...	51	117	S46.C	3	52	52	107
S46.H	62	...	60	122	S46.D	4	61	52	117
S46.J	52	...	46	98	S46.E	2	63	51	116
S46.K	71	...	46	117	S46.F	3	58	56	117
S46.L	60	...	51	111	S46.I	4	46	63	113
	363	...	319	682		24	329	336	689

The total absence of Mids in six of these progenies indicates, not only that the Short parents did not carry the mid gene, but also that little stray pollen was present at the crossing plot, nor did illegitimate unions take place between the Shorts, since the other six plants evidently do carry the mid gene.

In 1947 three of the Shorts carrying the mid gene were crossed with their Mid daughters, which must be heterozygous for Mid (Mm) as their pollen parent was a Long. Table 2 shows the classification of the progenies of the three Shorts and the mixed seed from their Mid daughters.

TABLE 2

Progenies from Ms/ms = Ms/mS *grown in 1948*

Plant Number	L	M	S	Total
S46.D	2	59	59	120
S46.E	4	59	57	120
S46.F	4	66	49	119
Mids	2	63	54	119
	12	247	219	478

If inheritance is disomic, the Mid offspring from such a cross (SsMm = ssMm) are expected to be either homozygous (MM) or heterozygous (Mm), and when backcrossed to Long, their progenies would be of two types—one segregating 1 Long : 1 Mid and the other consisting entirely of Mids. Whereas if tetrasomy existed, the genotype of the six mid-carrying Shorts shown in table 1 would be Ss_3Mm_3 and their Mid daughters—s_4Mm_3. The Mids resulting from an intercross of these two genotypes would be duplex—M_2m_2, and simplex—Mm_3, in the proportion of 1 : 2, and when backcrossed to Long, the duplex would segregate 5 Mids : 1 Long, while the simplex would give 1 M : 1 L. A third class might occur by double reduction, as was the case in *L. salicaria* (Fisher and Mather, 1943), when a triplex Mid—M_3m appeared. When this plant was backcrossed, almost all its offspring were Mids, the Longs that appeared being due to double reduction. It is apparent, then, that only when inheritance is disomic can any considerable fraction of the progenies of Mids obtained from the intercross described above be expected to contain only Mids.

Twelve Mids, obtained from the intercross of S46.D, S46.E and S46.F with their Mid daughters, were backcrossed to Longs, and in 1949 progenies of 30 were grown and classified for style length. As will be seen from table 3, seven gave no Longs and are presumably homozygous, while the other five segregated 1 : 1.

TABLE 3

Test of 12 Mids in 1949

Plant Number	L	M	Total	Plant Number	L	M	Total
M48.73	13	17	30	M48.72	...	28	28
M48.76	15	15	30	M48.74	...	30	30
M48.78	17	13	30	M48.75	...	30	30
M48.81	17	13	30	M48.77	...	30	30
M48.82	19	11	30	M48.79	...	30	30
				M48.80	...	30	30
				M48.83	...	30	30
5 plants	81	69	150	7 plants	...	208	208

LINKAGE OF MID AND SHORT LOCI

It will be seen from table 1 that, in the six progenies containing Mids, the ratio of Mid to Long is not the 1 : 1 expected with independence. All six progenies are, however, homogeneous, $\chi^2 = 6{\cdot}77$ for 5 degrees of freedom. The short to non-short ratio is homogeneous—$\chi^2 = 4{\cdot}72$ for 5 d.f. The simplest explanation for this unexpected ratio is that the mid and short loci are linked and that these six Short plants are all in repulsion—Sm/sM, which when backcrossed to Longs are expected to give mostly Shorts and Mids. Any Longs would be recombinants. The other recombinant class would be short-styled plants carrying the mid gene in coupling—SM/sm. Since the proportion of Longs is 6·79 per cent. of the non-shorts, a similar proportion, *i.e.* 1 in 15, of the Shorts can be expected to be of the genotype SM/sm. It was, therefore, decided to test 15 Shorts from these progenies by backcrossing to Longs, and in 1948, progenies of 100 from each Short were grown and scored for style length.

The results obtained are given in table 4. Fourteen progenies gave no Mids. S47.4, however, gave 2 Mids out of 27 non-shorts. The following year a progeny of 995 was grown from this plant. 528 Longs, 23 Mids and 444 Shorts were obtained. The proportion of Mids in the non-shorts represents the recombination fraction, and is rather less than was observed in the six progenies grown in 1947.

TABLE 4

Test of 15 Shorts in 1948

Plant Number	L	M	S	Total	Plant Number	L	M	S	Total
S47.4	25	2	38	65	S47.1	54	...	45	99
					S47.2	54	...	46	100
					S47.3	50	...	50	100
					S47.5	39	...	61	100
					S47.6	57	...	43	100
					S47.7	56	...	44	100
					S47.8	45	...	55	100
					S47.9	46	...	55	101
					S47.10	50	...	50	100
					S47.11	51	...	48	99
					S47.12	53	...	48	101
					S47.13	56	...	42	98
					S47.14	60	...	39	99
					S47.15	51	...	49	100
1 plant	25	2	38	65	14 plants	722	...	675	1397

Using the method of maximum likelihood, the estimate of the recombination fraction, obtained from all the data, is 5·74 per cent. The amount of information is 19832·6 from which is derived a standard error of 0·71 per cent. On the whole this is an estimate of recombination in the ovules, since the only pollen mother cells tested were of the Mids in table 2, which contributed little to the estimate.

FREQUENCIES OF THE 7 GENOTYPES

Short plants are always heterozygous in respect of the short allele, as a homozygote would only be produced by an illegitimate selfing or mating of Short with Short. In respect of their Mid content, however, Shorts may be any one of four genotypes. Mids are of two genotypes, and Longs of only one, the double recessive.

Assuming that the three forms are present in equal numbers in a population, with equal viability and fertility, the seven genotypic frequencies, when in equilibrium, would be—

Longs	Mids	Shorts
ms/ms 100 per cent.		mS/ms 53·6 per cent.
	Ms/ms 92·8 per cent.	MS/ms 19·6 ,,
		Ms/mS 19·6 ,,
	Ms/Ms 7·2 ,,	MS/Ms 7·2 ,,

COMPARISON WITH *LYTHRUM*

The striking similarities in the hereditary mechanism of tristyly in *Lythrum salicaria* and *Oxalis valdiviensis* are that the long-styled form is the double recessive, and that the mid gene is hypostatic to the Short. The differences are that *L. salicaria* is an autotetraploid with 60 chromosomes, forming some quadrivalents at meiosis, and showing tetrasomic inheritance, whereas *O. valdiviensis* is a diploid forming 9 bivalents and showing disomic inheritance : that the mid and short loci are independent in the former and linked in the latter. Fisher (1949) has shown that this linkage confers a slight advantage in that it makes inbreeding by sib-matings a little less likely to occur ; but it does not reduce the frequency of parent-offspring matings.

In *Lythrum* there has been a regular excess of Mids in progenies where 1 Mid to 1 Long is expected. In the *Oxalis* progenies containing short-styled plants, it will be seen that out of a total of 4306 plants only 2030 are Shorts, where half are expected. Although the deficiency of Shorts is less than 6 per cent., it is significant $-\chi^2 = 14{\cdot}05$ for 1 degree of freedom. All except 119 of these plants had Short mothers, so the possibility that this is a reciprocal difference cannot be ruled out. The 119 plants with Mid mothers (table 2) show a 4 per cent. deficiency of Shorts, but this is not significantly different from the 1 : 1 expected. After discussing this with Dr A. R. G. Owen, it became apparent that the observed deficiency would be quite compatible with a situation in which Ss zygotes had a viability of only 94 per cent. relative to ss zygotes. On account of the compulsory crossbreeding, such a disadvantage of one genotype relative to the other would not lead to the extinction of one of the alleles, and might well, in a wild population, lead to a stable proportion of these genotypes in the same ratio, *i.e.* of the viability.

In making this comparison, it is as well to note, however, that,

with the exception of the parent-offspring mating of three Shorts to their Mid daughters (table 2), no appreciable inbreeding has been done during the work on *Oxalis*, whereas, in *Lythrum* there was a considerable amount of inbreeding.

The genus *Oxalis* is a large one. In his taxonomic revision of the genus, Salter (1944) describes over 200 species in South Africa alone. Since almost all of them are tristylic, it would seem reasonable to assume that tristyly developed in their common ancestor rather than in each species independently or even in each group of species. They have, however, various chromosome numbers ranging from 10 to 80 —Darlington and Janaki Ammal (1945) give the basic numbers 5, 6, 7, 9, and 11—many of them are presumably polyploids. It is hoped that the experimental work may be extended on other species of *Oxalis*.

SUMMARY

1. Tristyly is a crossbreeding system. A knowledge of the inheritance of the long-, mid- and short-styled forms is required to show how this system works.

2. That the long form is recessive was shown by Darwin and confirmed by Barlow, who based her experiments on the assumption of a short gene and a mid gene, the former being epistatic to the latter.

3. In this investigation of the situation in *Oxalis valdiviensis* the open pollination method of Fisher and Mather was used in all crosses. Contamination and illegitimate unions were so rare that none was detected.

4. Twelve Shorts were crossed to Longs. Six of them were found to be carrying the mid gene. Three of these mid-carrying Shorts were then crossed to their Mid daughters, which were necessarily heterozygotes. Twelve Mids resulting from this intercross were tested by backcrossing to Longs. Five segregated 1 Long : 1 Mid, and seven gave no Longs and must, therefore, be homozygous Mids, thus showing that the inheritance of Mid is disomic.

5. The 6 mid-carrying Shorts detected from the first cross gave satisfactory 1 Short : 1 Non-short ratios. The Long : Mid ratio was 1 : 14 with a homogeneity χ^2 of 4·72 for 5 d.f., suggesting linkage of the mid and short loci, with all six Shorts in repulsion. Half the recombinants would be Longs, the other half would be Shorts carrying the mid gene in coupling. Fifteen Shorts were, therefore, tested by backcrossing to Longs. Fourteen gave no Mids in their progenies, but one gave 2 Mids out of 27 non-shorts, and in the following year, 23 out of 551 non-shorts.

6. The recombination fraction estimated by maximum likelihood on all the data is 5·74 per cent. with standard error 0·71 per cent. This estimate is based mainly on the recombination in the ovules.

7. Taking into account coupling and repulsion, there are seven genotypes. Four are phenotypically Shorts, two are Mids and one Long. The frequencies of the seven genotypes are given, assuming equal numbers of each phenotype with equal viability and fertility.

8. A comparison is made with the similar tristyly in *L. salicaria*.

I am greatly indebted to Professor R. A. Fisher for his collaboration throughout these experiments.

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LINKAGE BETWEEN FIDGET AND AGOUTI IN THE HOUSE MOUSE

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INTRODUCTION

THE recessive gene fidget in the mouse (symbol *fi*; Grüneberg, 1943) influences behaviour (head shaking, circling), the cornea and the skeleton (polydactylism and some other effects not yet described). Linkage tests of *fi* with tan, a^t, albinism, *c*, and blue dilution, *d*, all gave negative results; there was no close linkage, but the possibility of loose linkage could not be excluded.

During 1949 further breeding records led us independently to suspect loose linkage between *fi* and a^t. The combined results, which are presented in this paper, establish its existence; recombination is about 33·5 per cent. Work is now proceeding to determine the position of *fi* with respect to that of pallid, *pa*, which is also linked to a^t (Roberts and Quisenberry, 1935).

MATERIAL AND DATA

Two types of mating were used, namely repulsion intercrosses $a+/a^tfi \times a+/a^tfi$ and coupling intercrosses $A+/a^tfi \times A+/a^tfi$; but the progeny of one set of matings of the latter type were divided into three phenotypes with respect to the agouti locus, based on the appearance of the belly (light versus dark) as well as the back (agouti versus black).

The data are shown in table 1, which also reproduces those published previously (Grüneberg, 1943).

Several other mutants were also present in some of the matings; they were brown, *b*, albinism, *c*, luxate, *lx*, pinkeyed dilution, *p*, and macrocytic anæmia, W^v. It was not possible to classify fully for pinkeyed dilution, owing to the simultaneous segregation of albinism; nor was it possible to put any confidence in the classification of luxate, since the double heterozygote $+fi+lx$ was found to resemble the homozygote *lxlx* and the heterozygote $+lx$ often shows polydactyly similar to that seen in many fidgets. The segregations of *fi* with *b*, *c*, *p* and W^v are shown in table 3.

TABLE I

Segregation of fi *and* a^t

Author and date	Mating *	Phenotypes and numbers of progeny							Segregation of *fi* †					Segregation of a^t † ‡				
								Total	*D*	*I*	χ^2	*DF*	*P*	*D*	*I*	χ^2	*DF*	*P*
Edinburgh :—		a^t+	$a^t fi$	$a+$	$a\,fi$													
Falconer, new data	I.R.	21	10	16	...	...	...	47	$-9{\cdot}\dot{3}$	$250{\cdot}\dot{6}$	0·348	...	...	$-22{\cdot}\dot{6}$	$150{\cdot}\dot{6}$	2·050	...	...
Carter, new data	I.R.	34	13	16	3	...	...	66	$-2{\cdot}\dot{6}$	352·0	0·020	...	...	$-13{\cdot}\dot{3}$	352·0	0·505	...	...
		$AA+$	$AA\,fi$	Aa^t+	$Aa^t\,fi$	a^ta^t+	$a^ta^t\,fi$											
Carter, new data	I.C.	16	2	45	13	16	10	102	$-2{\cdot}\dot{6}$	544·0	0·013	...	...	+32·0	816·0	1·255	...	...
Sum . .	...	...	...	...	...	...	...	...	...	...	0·381	...	...	...	...	3·810	...	...
Deviation .	...	...	...	...	...	...	...	...	$-14{\cdot}\dot{6}$	$1146{\cdot}\dot{6}$	0·215	1	>0·5	−4·0	$1418{\cdot}\dot{6}$	0·011	1	>0·9
Heterogeneity .	...	...	...	...	...	...	...	...	...	...	0·166	2	>0·9	...	...	3·799	2	>0·1
London :—		a^t+	$a^t\,fi$	$a+$	$a\,fi$													
Grüneberg, new data	I.R.	226	75	105	6	...	...	412	$-117{\cdot}\dot{3}$	$2197{\cdot}\dot{3}$	6·265	...	...	$-42{\cdot}\dot{6}$	$2197{\cdot}\dot{3}$	0·828	...	...
		$A+$	$A\,fi$	a^t+	$a^t\,fi$													
Grüneberg, 1943	I.C.	53	11	21	7	...	...	92	$-26{\cdot}\dot{6}$	$490{\cdot}\dot{6}$	1·449	...	...	$+13{\cdot}\dot{3}$	$490{\cdot}\dot{6}$	0·362	...	...
Sum . .	...	...	...	...	...	...	...	...	...	...	7·714	...	...	...	...	1·191	...	...
Deviation .	...	...	...	...	...	...	...	...	−144·0	2688·0	7·714	1	<0·01	$-28{\cdot}\dot{3}$	2688·0	0·299	1	>0·5
Heterogeneity .	...	...	...	...	...	...	...	...	...	...	0·000	1	>0·9	...	...	0·892	1	>0·3

* In the column headed " mating," C = coupling, I = intercross, R = repulsion.

† In the segregation analyses, *D* = discrepancy in the equation of estimation, *DF*= degrees of freedom, *I* = amount of information, *P* = probability.

‡ In all cases a^t entered with *fi* ; the signs have, therefore, been taken in the sense required to test the segregation of a^t from its allele, whether a^t was playing the dominant or recessive role.

ANALYSIS OF THE DATA

The data have been analysed by the methods described by Mather (1935, 1937), which are special applications of the maximum likelihood method of estimation (Fisher, 1925). In the linkage analysis, however, we have adopted the sign convention used by Fisher (1946), whereby linkage closer than the trial value is indicated by a positive instead of a negative score.

The single-factor segregation analyses (table 1) show a deficiency of fidgets, significant at the 1 per cent. level, in the London data ;

TABLE 2

Linkage of fi *and* a^t

Author and date	Mating	Analysis of linkage					
		D_{23}	D_{24}	I	χ^2	DF	P
Edinburgh :—							
Falconer, new data .	I.R.	+12·685	+13·241	55·613	2·893	...	...
Carter, new data .	I.R.	−7·343	−6·278	106·554	0·506	...	...
,, ,, .	I.C.	−9·672	−6·552	311·996	0·300	...	...
London :—							
Grüneberg, new data	I.R.	+26·266	+30·463	419·738	1·644	...	...
,, ,, 1943 .	I.C.	−27·912	−24·904	300·726	2·591	...	...
Sum . . .	...	...	...	...	7·934	...	...
Deviation . .	...	−5·976	+5·970	1194·628	0·030	1	>0·9
Heterogeneity .	...	...	...	...	7·904	4	>0·05

Recombination fraction $p = 33{\cdot}5 \pm 2{\cdot}9$ per cent. (standard error).

TABLE 3

Segregation of fi *with* b, c, p, W^v

Author	Mates	Phenotypes and numbers of progeny						
				++	+*fi*	*b* +	*b fi*	Total
Grüneberg	*b*+/+*fi*×*b*+/*b fi*	...	...	23	5	33	8	69
,,	*b*+/+*fi*×*b*+/+*fi*	...	...	84	29	30	7	150
				++	+*fi*	*c* +	*c fi*	Total
Falconer	*c fi*/++×*c fi*/*c fi*	...	...	19	3	16	10	48
,,	*c fi*/++×*c fi*/++	...	...	85	22	26	8	141
,,	*c fi*/++×*c fi*/*c*+	...	...	6	2	5	3	16
Grüneberg, new data	+*c fi*/*p*++×+*c fi*/*p*++	...	...	331 *112**	82 *35**	116	29	558
		W^vW^v +	W^vW^v*fi*	W^v+ +	W^v+*fi*	++ +	++*fi*	Total
Falconer	W^v+/+*fi*×++/+*fi*	...	...	11	2	11	4	28
Carter	W^v+/+*fi*×W^v+/+*fi*	8	...	20	5	14	5	52

* Figures in italics show the numbers of pinkeyed progeny within the non-albino classes.

the deficiency of a^t in the London data is not significant and the Edinburgh data show good segregations at both loci.

In analysing the linkage we assume that the deficiency of fidgets was entirely due to inviability of the homozygotes and that this

inviability was not dependent upon the genotype at the agouti locus. When these assumptions are made, the estimate of the recombination fraction is independent of the viability of the fidgets : the amount of statistical information available about the estimate is still measured by the rate of change of the discrepancy in the equation of estimation.

The linkage analysis is shown in table 2 : the five bodies of data are homogeneous and indicate linkage with a recombination fraction $33{\cdot}5 \pm 2{\cdot}9$ per cent. (standard error).

The data of table 3, which gives the segregations of *fi* with *b*, *c*, *p* and W^v, do not show any significant departures from free segregation.

SUMMARY

Fidget, *fi*, in the house mouse is linked to tan, a^t ; recombination is estimated at $33{\cdot}5 \pm 2{\cdot}9$ per cent. Its position with respect to pallid, *pa*, is not yet known.

Acknowledgments.—We are grateful to Dr D. S. Falconer for permission to quote some data from the Animal Breeding and Genetics Research Organisation's laboratory at the Institute of Animal Genetics, Edinburgh. One of us (H. G.) was assisted by a research grant from the Medical Research Council.

Note added 2nd January 1950.—We are grateful to Professor Fisher for pointing out, since this paper was written, that the linkage analysis of some of our data (the four class segregations) could have been facilitated by the use of table XIV_1, in the 1948 edition of Fisher and Yates' *Statistical Tables for Biological, Agricultural and Medical Research* (Edinburgh, Oliver and Boyd).

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THE BLOOD GROUPS OF THE PEOPLE OF EGYPT

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By means of its land connection with Asia, Egypt is one of the gateways through which large movements of people into and out of Africa have taken place. Physical anthropologists nevertheless point to a general persistence of physical type in Egypt throughout many thousands of years. A comparison of the blood groups of the people of Egypt with those of their neighbours must therefore be of great interest. Numerous studies of their ABO and MN groups, summarised by Boyd (1939), had already been made but nothing was known of their Rh groups.

TABLE 1

The A_1A_2BO groups of 144 Egyptians

Group	Moslems	Christians	Total observed	Number expected	Frequency observed
O	23	16	39	*42·1*	0·271
A_1	16	28	44	*42·2*	0·306
A_2	4	1	5	*4·4*	0·035
B	27	17	44	*40·1*	0·306
A_1B	4	7	11	*13·4*	0·076
A_2B	0	1	1	*1·7*	0·007
Total	74	70	144	*143·9*	1·001

Gene frequencies

O	0·541
A_1	0·217
A_2	0·027
B	0·215
	1·000

It is well known that the present population of Egypt is markedly heterogeneous. The educated classes in particular have received recent accessions from various other countries, especially around the eastern Mediterranean; in the opinion of some anthropologists one of the oldest established elements in the population, and one bearing a close resemblance in physical characters to the ancient Egyptians of Pharaonic times, consists of those people in Upper Egypt who,

TABLE 2

The MNS *groups of 144 Egyptians*

Group	Moslems	Christians	Total observed	Number expected	Frequency observed
MS	11	16	27 } 39	25·7 } 38·1	0·187
Ms	4	8	12 }	12·4 }	0·083
MNS	20	17	37 } 70	36·6 } 71·9	0·257
MNs	16	17	33 }	35·3 }	0·229
NS	6	3	9 } 35	8·8 } 34·1	0·062
Ns	17	9	26 }	25·3 }	0·181
Total	74	70	144	144·1	0·999

Chromosome frequencies

MS	0·221 }	0·514
Ms	0·293 }	
NS	0·067 }	0·486
Ns	0·419 }	
Total	1·000	

TABLE 3

The Rh *groups of 184 Egyptians*

Phenotype	Commonest genotype	Number observed	Number expected	Frequency observed
CCDee *	CDe/CDe	47	45·0	0·255
CcDee	CDe/cde	73	75·7	0·397
CcDEe	CDe/cDE	15	16·3	0·081
ccDEE	cDE/cDE	1	1·5	0·005
ccDEe	cDE/cde	16	13·7	0·087
ccDee	cDe/cde	20	19·9	0·109
ccD^{u}ee	cD^{u}e/cde	1·09 †	1·1	0·006
ccddee	cde/cde	10·91 †	10·8	0·059
Totals		184	184·0	0·999

* See Mourant (1949).

† Out of 12 samples found on preliminary testing to be cde/cde, 11 were further tested for the presence of the D^{u} antigen which was found in one of them. The remaining one had been tested before the test for D^{u} had become standard practice. It has therefore been partitioned between the two phenotypes in the proportion of 10 : 1.

Chromosome frequencies

CDe *	0·495
cDE	0·090
cDe	0·161
cD^{u}e	0·012
cde	0·243
Total	1·001

* The anti-C serum used had the specificity anti-C+anti-C^{w}. CDe therefore includes any C^{w}De which may have been present in the population.

at the present time, practise the rites of the Coptic Christian Church. The heterogeneity of modern Egyptians is reflected in the variations in the frequencies of the ABO blood groups.

Whilst we should have preferred to confine our studies to one or more groups of people selected on anthropological grounds we have hitherto been unable to obtain sufficient suitable specimens for this purpose : and the results obtained by examining specimens obtained through a variety of channels and selected in different ways show, nevertheless, sufficient homogeneity from the blood group point of view and at the same time are of such great interest and show such a great difference from all other populations so far examined, as to make it desirable to publish this account at once rather than to delay in order to add to our data. We hope that we ourselves and other workers will later be able to supplement these by many more tests.

In 1947 Dr R. R. Race determined the ABO and Rh groups of 20 Moslems and 20 Coptic Christians of Egypt, from specimens of blood kindly sent through the courtesy of Brigadier H. T. Findlay. Dr Race has kindly allowed us to use these results. Subsequently one of us (K. A. I.) was able to collect specimens from a considerable number of Egyptians resident in London. Then we received from Professor J. H. Fisher two consignments of specimens obtained from Coptic Christians of Upper Egyptian origin. These were specially selected from the anthropological point of view and as far as could be ascertained by careful questioning, none of the subjects had any known ancestors born outside Egypt. The numbers of them are too small at present for separate statistical treatment, but to enable them to be used in any future study in combination with results obtained from other people selected in a similar manner, their full blood groups are listed in table 4. The remaining Christians tested are probably comparable but we have no guarantee of this other than the similarity of their blood-group frequencies.

A small number of the Moslems are known to have had some non-Egyptian ancestors, but an attempt to eliminate such persons from our lists would have been very difficult and liable to lead to errors due to bias arising from the known results of the tests, errors possibly greater than have been introduced by their retention. We thus had available altogether the results of testing 184 persons, 94 Moslems and 90 Christians. All of these were tested for Rh (with anti-C+anti-C^w, anti-D, anti-E, anti-c and anti-e). In the first 40 tests carried out by Dr Race no distinction was possible, owing to the age of the specimens, between A_1 and A_2; since the total frequencies of the groups O, A, B, and AB among these 40 differed little from those found in the subsequent 144, the former have been disregarded in our ABO calculations. The first 40 were also not tested for MN and S. Among all but the same 40, apparent D-negatives were subjected to an indirect Coombs test with a strong incomplete anti-D serum in order to detect the D^u antigen if present.

From our results we have calculated gene frequencies by methods already used by some of us (Chalmers, Ikin and Mourant, 1949; Prasad, Ikin and Mourant, 1949) and from these the expected phenotype numbers have been calculated and compared with the observed values. The Rh calculations are, however, somewhat complicated by the fact that the group ccee here includes three phenotypes, ccDee, ccD"ee and ccddee. The chromosome frequencies cDe, cD"e and cde were therefore calculated from the frequencies of these phenotypes and a proportional correction made so that the sum of

TABLE 4

Sixty-two anthropologically selected Copts of Upper Egyptian ancestry classified according to A_1A_2BO *and* MNS *groups and probable* Rh *genotypes*

Probable Rh genotype	AB	A_1	B	O
CDe/CDe	Ms 1 MNS 1 * MNs 1	Ms 1 MNS 2 MNs 1 Ns 1	MS 1 MNS 1 MNs 2	MS 1 MNS 1 MNs 1
CDe/cDE	...	MNS 1 MNs 2	...	...
CDe/cde	MS 1 MNs 1	MS 2 Ms 3 MNS 3 MNs 3	MS 1 Ms 3 MNS 1 MNs 1	MNS 2 NS 2 Ns 1
cDE/cde	MNS 1	...	MS 1 MNs 1 Ns 1	MS 2 MNs 1
cDe/cde	Ns 1	MS 2 Ns 1	Ns 1	MS 1 MNS 1 MNs 1
cD"e/cde	...	...	MNs 1	...
cde/cde	...	Ns 2	MNS 1	MS 1

* All the persons classified as AB were A_1B with the exception of this one who was A_2B.

the chromosome frequencies should agree with the total frequency of ce chromosomes found by the methods described by Chalmers, Ikin and Mourant (1949). These methods are adopted as expeditious though known not to be fully efficient.

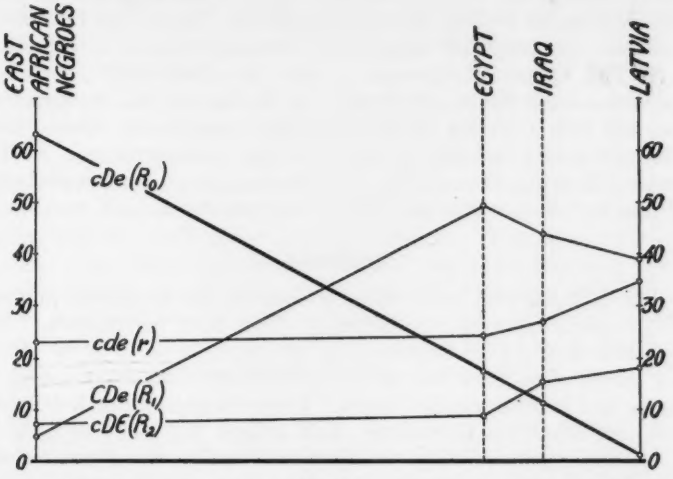
We have shown in tables 1 and 2 separate totals of Christians and Moslems but have based all our calculations on the totals of both. The differences in Rh phenotype frequencies are not given because they were (table 3) well below the level of significance. Even in the case of ABO and MN, where they are greater, they are still not significant.

We do not propose in this paper to undertake a critical study of

the ABO and MN groups of the population of Egypt. Among the wide range of frequencies found by different observers and quoted by Boyd (1939) it is difficult to select the most reliable but it is clear that our results do not differ greatly from those of most previous workers. As a rough estimate the average percentage gene frequencies found by others among Christians are A, 25 ; B, 21 ; O, 54 ; and

Rh CHROMOSOME FREQUENCIES (PER CENT) IN EAST AFRICA, WESTERN ASIA AND EASTERN EUROPE.

Based on the present study, on the work of Kayssi (1949), Race, Sanger, Lawler and Keetch (1948), and on unpublished data of Ikin and Mourant.



The diagram shows the fall in frequency of the cDe chromosome from south to north, the maximum frequency of CDe in Egypt, and the corresponding variations in frequency of the other common Rh chromosomes. The spacing on the horizontal scale is arbitrarily arranged so as to make the frequencies of cDe fall on a straight line.

FIG. 1.

among Moslems A, 28 ; B, 21 ; O, 51 ; whereas our overall figures show A, 24 ; B, 21 ; O, 54, with, in fact, a higher frequency of A among Christians than among Moslems. We have found the ratio of the phenotype frequencies A_2/A_1, to be 0·11 which is lower than the values (0·17 to 0·24) found by previous workers and quoted by Boyd (1939). From Boyd's compilation of MN data the gene percentages for both Moslems and Christians are roughly M, 53 ;

N, 47 whereas we find M, 51 ; N, 49. Our determinations of the Ss subgroups of the MN system are the first to be published for Egyptians.

In their Rh phenotype frequencies the Egyptians differ very greatly from all other populations yet examined except the people of Baghdad (Kayssi, 1949) to whom they show a moderate degree of resemblance. The outstanding feature of the Egyptians is their very high CDe(R_1) frequency, considerably higher than that of any population in Europe for which figures have been published, and very much higher than some of us have found in testing numerous other African populations (Donegani, Ikin and Mourant, unpublished). At the same time they show a higher cDe(R_0) frequency than any non-Negro population nearer than Eastern Asia. While any final conclusion must depend on the examination of many other Near Eastern populations, an examination of fig. 1 in which the Rh chromosome frequencies of East Africans, Egyptians, Iraqui and Europeans (Latvians) are compared suggests the following tentative conclusions :

(i) The Egyptians represent a basic or culminating population type with a small Negro admixture. (ii) In Iraq we find the Egyptian type, still with a certain Negro admixture, considerably diluted with European stock ; indeed, in view of recent observations on the Rh groups of Indians (Prasad, Ikin and Mourant, 1949) we can probably say that the dilution was with Indo-European stock.

SUMMARY

The A_1A_2BO and MNS blood groups of 144 Egyptians, and the full Rh phenotypes of 184 Egyptians have been determined. The Egyptians show a high frequency (21 per cent.) of the B gene. Their MN distribution does not differ significantly from that found in Europe and in other parts of Africa. Their Rh distribution is different from any other yet discovered, with a high frequency of CDe or R_1 (49 per cent.) which is probably an ancient Egyptian characteristic, and with 16 per cent. of cDe(R_0) chromosomes, a feature probably derived from an admixture of Negro stock.

Acknowledgments.—We wish to thank Brigadier H. T. Findlay, Prof. J. H. Fisher, Dr J. N. Marshall Chalmers and many other persons who helped in the collection and transport of the specimens for this work ; also Dr R. R. Race for allowing us to use the results of his tests on 40 Egyptians.

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THE *DUFFY* BLOOD GROUP SYSTEM

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DURING the investigation of a hæmolytic transfusion reaction it was observed that the patient's serum contained an unusual antibody; tests against a large number of human bloods showed that the factor thus defined was one which had not previously been recognised. A brief account of these observations has already been published (Cutbush, Mollison and Parkin, 1950). The purpose of the present paper is to give a fuller account of the investigations and to present some additional observations.

CASE HISTORY

The patient (R. D.), aged 43, was a sufferer from hæmophilia. Signs of the disease were first manifest in infancy and there had since been many attacks of spontaneous bleeding. Up to July 1949 the patient had had only three blood transfusions. The first was given in 1928, when persistent bleeding followed the extraction of a tooth. This transfusion was followed by a rigor. The patient received his second and third transfusions in 1936: both were followed by rigors but no further details are available. One week before the first of these two transfusions the patient received an intramuscular injection of blood.

The patient then remained in moderately good health until 7th July 1949, when he was admitted to hospital with spontaneous bruising and bleeding. On 11th July, the patient was in need of a transfusion; he was found to be group *O Rh* negative and the blood of 3 group *O Rh* negative donors was tested against his serum as follows: The cells of each donor, suspended in 20 per cent. albumin, were mixed with an equal volume of the patient's serum; the mixtures were left at 37° C. for an hour and the sediments were then examined microscopically. No trace of agglutination was observed in any of the three mixtures. The transfusion was started and, although the patient developed a rigor during its course, the blood from all three donors was eventually given.

On the following day the patient became jaundiced and further investigations were therefore undertaken.

It was now found that using the indirect Coombs test, the patient's serum reacted with the blood of the first and third donors, though not with the blood of the second donor. The fate of the cells from the third donor could be followed, since the donor belonged to

type *N* and the patient and the other donors to type *MN*. A test with a potent anti-*M* serum, 48 hours after transfusion, showed that there were only some 60,000 type *N* cells per c.mm. in the patient's circulation, compared with the expected number of approximately 400,000 per c.mm.

Four days later a further transfusion was given. A donor was selected from *Rh* negative persons whose cells gave a negative Coombs test after incubation with the patient's serum. This transfusion was not accompanied by any unfavourable reaction. Another successful transfusion was given 7 days later, and soon afterwards the patient was able to leave hospital.

EXAMINATION OF THE SERUM

A sample of serum, taken before the incompatible transfusion, was found to contain anti-*Rh* and a second antibody which reacted with a large proportion of *Rh* negative bloods. A further sample, taken 11 days after the incompatible transfusion, reacted with the same bloods as before but the reactions were now considerably stronger.

It has already been mentioned that the pre-transfusion sample of serum failed to agglutinate suspensions of the donors' cells in 20 per cent. albumin, although it reacted with 2 of the 3 donor bloods when the indirect Coombs test was used.

Samples of serum were obtained at intervals after transfusion and tube tests were carried out as follows : Serial dilutions of the serum in saline and in albumin were mixed with equal volumes of group *O Rh* negative cells suspended in saline and albumin respectively. The mixtures were left for 2 hours at 4° C., 22° C. and 37° C., and the sediments were than examined microscopically. In a few instances weak rouleaux formation was observed in the first and second tubes of the titration but definite agglutination was never observed. However, the same cells, after incubation with the patient's serum for 30 minutes at 37° C., followed by three washings in saline, were strongly agglutinated by an anti-human-globulin serum.

Further tests were made, using cells which had been incubated at 37° C. for 1 hour with a buffered solution of trypsin, and then washed three times. Suspensions of these cells were mixed with serial dilutions of the patient's serum in saline and in albumin. Weak agglutination was observed in the first three tubes of the titration in saline and in albumin, but the reactions were no stronger with *Duffy* positive cells (*i.e.* those giving a positive indirect Coombs test) than with *Duffy* negative cells.

SEPARATION OF THE NEW ANTIBODY

All the samples of serum examined contained four antibodies, namely anti-*A*, anti-*B*, anti-*D* and the antibody which reacted with

certain group *O Rh* negative bloods. The antibody responsible for these latter reactions was isolated as follows : The serum was incubated at 37° C. with an equal volume of washed *O Rh* negative cells which had previously been shown to react with the serum, using the indirect Coombs test. The cells were then washed 3 times in saline at 37° C. An equal volume of saline was added to the packed cells and the mixture was heated at 56° C. for 10 minutes, then immediately centrifuged in buckets containing water at 56° C. The supernatant fluid was found to react with the same *O Rh* negative bloods as did the original serum. Although the reactions were now slightly weaker, this eluate could be used to test all cells irrespective of their *ABO* group or *Rh* type. It was thus possible to select *AB Rh* positive cells which did not react with the eluted antibody and to use these cells for absorbing the original serum. Two absorptions with equal volumes of cells were found to be sufficient to remove anti-*A*, anti-*B* and anti-*D* from the original serum and thus to prepare a serum containing the *Duffy* antibody alone.

REACTIONS OF THE *DUFFY* SERUM

The absorbed serum was used to test a large number of blood samples, in parallel with other available blood group sera. In table 1 the reactions of a sufficient number of bloods are set out to

TABLE 1

	Duffy	*ABO*	*Rh*	*MN*	*S*	*P*	*Kell*	*Lutheran*	*Lewis*	Secretor	P.T.C.
R. D. (patient) .	−	*O*	*rr*	*MN*	+	+	−	−	*a*− *b*+*	+	...
Donor i . .	+	*O*	*rr*	*MN*	+	...	−	...	*a*+ *b*−	...	...
Donor iii . .	+	*O*	*rr*	*N*	−	+	+	...	*a*− *b*+	...	...
C. M. . .	+	*O*	*rr*	*M*	+	+	−	+	*a*− *b*+	+	+
M. P. A. . .	+	*O*	*rr*	*M*	+	+	−	−	*a*+ *b*−	−	−
V. E. . .	+	*O*	*rr*	*M*	+	−	−	−	*a*− *b*+	+	+
H. F. . .	+	*O*	R_1r	*N*	−	+	−	−	*a*− *b*+	+	−
M. C. . .	+	*O*	R_1R_1	*N*	−	+	−	+	*a*− *b*+	+	−
D. P. . .	+	*B*	*rr*	*M*	−	+	−	−	*a*−	+	+
N. P. . .	+	A_2B	R_1r	*MN*	−	−	−	−	*a*−	+	−
Donor ii . .	−	*O*	*rr*	*MN*	+	+	−	...	*a*− *b*+	...	...
M. E. H. . .	−	*O*	*rr*	*MN*	+	+	−	−	*a*+ *b*−	−	+
Ba. K. . .	−	*O*	R_1R_1	*N*	−	...	−	+	...	...	...
S. M. . .	−	*O*	R_1R_2	*M*	−	+	−	−	*a*−	+	+
Ba. B. . .	−	*A*	R_1r	*MN*	+	...	−	+	...	...	...
G. I. E. . .	−	A_1	R_1r	*MN*	−	+	+	−	*a*− *b*+	+	+
E. J. K. . .	−	A_1	R_1r	*MN*	+	−	−	−	*a*+	−	+
P. L. M. . .	−	A_2	R_1r	*MN*	+	+	−	−	*a*− *b*+	+	+

* Dr W. T. J. Morgan kindly tested the patient's saliva and demonstrated the presence in it of the Le^a substance. Thus the patient's genotype was Le^aLe^b.

demonstrate that the factor with which the serum reacts is not one of the antigens already recognised. Moreover, there is no obvious relationship to the ability to secrete *A*, *B* and *H* substances nor to the

ability to taste phenylthiocarbamide (P.T.C.) in a dilution of 1 in 20,000.

The serum has been tested against blood samples from 205 unrelated English adults of known *ABO* group. Of the 205 bloods, 133 (64·9 per cent.) gave a positive reaction. The incidence of positives among group *O* persons was not significantly different from that among persons belonging to groups *A*, *B* or *AB* (see table 2).

TABLE 2

ABO group	No.	Reaction with *Duffy* serum		
		Positive	Negative	Per cent. positive
O	123	77	46	*62·6*
A	72	48	24	*68·3*
B	6	4	2	
AB	4	4	0	
Total .	205	133	72	*64·9*

In 168 cases, the reaction of the blood sample with anti-*D* was also known. It appears that the reactions of anti-*D* and the *Duffy* antibody are independent of one another (see table 3).

TABLE 3

Reaction with anti-*D*	No.	Reaction with *Duffy* serum		
		Positive	Negative	Per cent. positive
Rh positive . .	87	56	31	*64·4*
Rh negative . .	81	51	30	*63·0*
Total . .	168	107	61	*63·7*

NOTATION

The system is conveniently described as the *Duffy* system. Evidence will be presented that the serum reacts with an antigen that is inherited, and that this antigen can be detected in the red cells of heterozygotes as well as homozygotes. It is proposed to call the antigen Fy^a; the same symbol may be used for the gene. Its hypothetical allelomorph may be called Fy^b. The present serum may be described as anti-Fy^a. The phenotype $Fy(a+)$ thus includes bloods of genotype Fy^aFy^a and Fy^aFy^b, and the phenotype $Fy(a-)$ only the genotype Fy^bFy^b.

From our data the following gene frequencies have been calculated :

$$Fy^a = 0{\cdot}4075 \quad Fy^b = 0{\cdot}5925$$

FAMILY INVESTIGATIONS

In 27 families, the blood of each parent and at least one child was tested against the anti-Fy^a serum. When one or both parents were $Fy(a+)$, 24 (66 per cent.) of the 36 children were $Fy(a+)$. In contrast, no $Fy(a+)$ children were found in families where both parents were $Fy(a-)$. This is convincing evidence that the Fy^a antigen is inherited.

In 3 families in which both parents were $Fy(a+)$, 4 of the children were $Fy(a-)$. Thus the phenotype $Fy(a+)$ must include bloods of genotype Fy^aFy^b.

From the known gene frequencies, the proportion of positive and negative children expected from the three distinguishable matings can be calculated.

It will be seen in table 4 that the observed figures are very close to those expected.

TABLE 4

Type of mating	Parents			Children				
	Proportion expected	No. expected	No. observed	Total	$Fy(a+)$		$Fy(a-)$	
					No. expected	No. observed	No. expected	No. observed
$Fy(a+) \times Fy(a+)$	*0·4210*	11·4	10	15	12·9	11	2·1	4
$Fy(a+) \times Fy(a-)$	*0·4557*	12·3	11	21	13·2	13	7·8	8
$Fy(a-) \times Fy(a-)$	*0·1233*	3·3	6*	13	0·0	0	13·0	13
Totals .	*1·0000*	27·0	27	49	26·1	24	22·9	25

* The number of $Fy(a-) \times Fy(a-)$ matings is greater than might be expected in a random sample. However, this can be accounted for by selection : when a child was found to be $Fy(a-)$, a special effort was made to obtain blood from the parents, for testing.

ADDITIONAL OBSERVATIONS

Saliva. An attempt was made to demonstrate the presence of the Fy^a substance in the saliva of $Fy(a+)$ persons, including some known to secrete *A*, *B* or *H* substances. Equal volumes of saliva and anti-Fy^a serum were mixed and allowed to stand at room temperature for one hour and then at 37° C. for half an hour. The mixture was then tested against known $Fy(a+)$ cells using the indirect Coombs test. The reactions were as strong as those given by controls to which no saliva had been added. Thus it was not possible by this method to detect Fy^a substance in saliva.

DEVELOPMENT OF THE *Fy*^{a} ANTIGEN

The blood of an embryo (crown-rump length 70 mm.) was tested and found to give a positive reaction with anti-*Fy*^{a} serum. The cord blood of 25 infants was tested for the presence of the *Fy*^{a} antigen; 18 (72 per cent.) of these samples gave a positive reaction. These reactions were no weaker than those given by adult bloods. It may be concluded that the *Fy*^{a} antigen is detectable at an early stage in fœtal life and is fully developed at birth.

THE *DUFFY* GROUPS OF A SAMPLE OF INDIAN PEOPLE

All the observations referred to so far were made upon samples of blood from English people. Through the kindness of Dr I. M. Chaudhri of the Blood Group Reference Laboratory, we were able to test blood samples from 55 Pakistani nationals, resident in London. Forty-seven of the subjects came from North-West India (Punjab, Frontier Province, Sind) and the remainder from other parts of India (5 from United Province, 3 from Madras or Bengal).

Fifty-one of the 55 samples, or 92·7 per cent., were *Fy*(*a*+):

Total	Area of India	*Fy*(*a*+)	*Fy*(*a*−)
47	North-West	43	4
8	Other parts	8	0
55	Total	51	4

It is possible that the incidence of the *Fy*^{a} antigen varies in different parts of India, and it may therefore be more useful to consider only the North-West Indian blood samples. The incidence of positive reactions amongst these was 91·5 per cent. (43 out of 47). This may be compared with the figure of 64·9 per cent. for the English samples. The observed difference is highly significant (more than 6 times the S.E. of the difference). The frequency of the *Fy*^{a} gene calculated from these figures is 0·708 for North-West India, compared with 0·413 in England.

This finding suggests that estimates of the incidence of the *Fy*^{a} gene in various populations may prove valuable in anthropological studies.

DISCUSSION

A sufficiently large number of blood samples has been tested to make it certain that the *Duffy* system is not very closely or absolutely linked either to the *ABO* or *Rh* systems. It remains possible that looser linkage to one of these two systems will be revealed by family

studies ; or that testing of large numbers of blood samples with the *Duffy* serum and with other rare sera, such as anti-*Kell*, will reveal some association with another blood group system.

In any case, whether the *Duffy* system proves to be entirely independent of the other systems, or related in some way to them, it appears that it will be a useful system from the point of view of genetical investigation, simply because both the distinguishable phenotypes are of frequent occurrence.

SUMMARY

1. An antibody, reacting with a hitherto undescribed antigen in human red cells, has been found in a sample of human serum. The antigen thus revealed is termed Fy^a and the same symbol is used for the gene ; the hypothetical allelomorph is called Fy^b. Samples of blood from 205 unrelated English people have been tested. The frequency of the phenotype $Fy(a+)$ was 64·9 per cent. The following gene frequencies have been calculated :

$$Fy^a = 0{\cdot}4075 \quad Fy^b = 0{\cdot}5925.$$

2. The serum had one interesting serological characteristic : it failed to agglutinate cells suspended in saline and albumin ; and the antibody could only be detected by the indirect Coombs test.

We should like to thank Dr Russell Fraser for permitting us to make observations on his patient ; the patient himself for permission to use his name to describe the blood group system revealed by his serum and for giving blood on several occasions ; and Dr R. R. Race for advice on the preparation of the paper.

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CUTBUSH, MARIE, MOLLISON, P. L., AND PARKIN, DOROTHY M. 1950. *Nature*, Lond., *165*, 188.

JAPANESE PAPERS ON GENETICS 1942-1948. ENGLISH ABSTRACTS

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I. PLANT CHROMOSOMES

HAGA, T. 1942. Geographical distribution of Trilliaceæ plants in relation to polyploidy. *Jap. Jour. Genet., 18,* 168-171.

Trillium kamtschaticum, a diploid ($2x = 10$), is distributed to the north up to northern Saghalien and Kamtschatka through Kurile islands, the southern limit terminating in the northern districts of Honshu. Distribution of tetraploid *T. Tschonoskii* ($4x = 20$) covers the area from southern Saghalien to Formosa. Sterile triploid ($3x = 15$) and fertile hexaploid form ($6x = 30$) of *T. Hagæ*, which are the hybrids between *T. kamtschaticum* and *T. Tschonoskii*, are found in the regions where the distributional areas of the above two species overlap. *T. Smallii*, a tetraploid ($4x = 20$), is recorded in middle Kuriles and middle Saghalien to the north and up to Shikoku to the south. *T. amabile*, a fertile hexaploid hybrid ($6x = 30$), probably, between *T. kamtschaticum* and *T. Smallii*, is revealed as occurring in the overlapping region of the distributions of the supposed parental species.

In Japan two species are known in the genus Paris, that is, *P. tetraphylla* and *P. verticillata*, both being diploid ($2x = 10$). Distribution of these two species similarly ranges from south Saghalien to Kyushu. *Kinugasa japonica*, an octoploid species ($8x = 40$), which shows morphological characteristics intermediate between *Trillium* and *Paris* is known to occur in the northern part of Honshu, where all the species of *Paris* and *Trillium* happen to inhabit.

HAGA, T. 1944. Morphological reaction of chromosomes to low temperature. *Jap. Jour. Genet., 20* : 87.

Patterns of differential segments were described for the somatic chromosomes of *Trillium kamtschaticum* which were chilled at 0° C. for 5 days. Size and position of the differential segments were found to be constant and homozygous for each chromosome pair, but different pairs showing different patterns. Taking the nucleus as a whole, all 13 plants sampled from a population of Siraoi, Hokkaido, were revealed to show one and the same pattern of differential segments, suggesting a highly homozygous condition of the present population.

HAGA, T., and KURABAYASHI, M. 1947. Genome analysis in the genus *Trillium* on the basis of differential reaction of the chromosomes. *Jap. Jour. Genet., 22,* 8.

On the basis of chromosome morphology, meiotic chromosome behaviour, external morphology and ecological findings, the genomic constitutions of five species of the genus *Trillium* known in Japan were concluded to be as shown in the following table.

It is revealed that the pattern of differential segments is characteristic for each genome. With this characteristic, constitutional genomes combined in the polyploid

species were identified with those genomes in diploid or in another polyploid, confirming the earlier conclusion reached from other lines of investigations.

	Chromosome number	Genomic constitutions
T. kamtschaticum . . .	$2x = 10$ 5_{II}	K_1K_1
T. Haga	$3x = 15$ $5_{II}+5_I$	K_1K_2T
T. Haga	$6x = 30$ 15_{II}	$K_1K_1K_2K_2TT$
T. Tschonoskii . . .	$4x = 20$ 10_{II}	K_2K_2TT
T. Smallii	$4x = 20$ 10_{II}	SSUU
T. amabile	$6x = 30$ 15_{II}	K_2K_2SSUU

HAGA, T., and KURABAYASHI, M. 1948. Chromosomal variation in *Trillium kamtschaticum*. *Jap. Jour. Genet.*, *23*, 12-13.

Chilled somatic chromosomes of 86 plants from 4 different populations of *Trillium kamtschaticum* were classified by the patterns of differential segments. There were identified 14 types in chromosome A, 7 in B, 6 in C, 8 in D and 4 in E. Knots and ring-fragments were observed at prophase of the haploid nuclei in the process of embryosac formation. Based on these aberrations, an interpretation was set forth, that, at least, some of the chromosomal variations may have arisen through inversions involving simultaneously both the normal and differential segments and deletion of the inert differential segments. Numbers of chromosome types found in the four populations are tabulated below. Some of the chromosome types were particular to a given population, but the majority of them were found in common through all four populations. However, the frequencies of some of the chromosome types were found to be different among the populations.

Chromosome	Siraoi	Sizunai	Samani	Akkesi
A	2	8	10	7
B	2	4	2	3
C	1	6	5	2
D	2	7	8	3
E	1	4	4	4
Total . . .	8	29	29	19
No. of plants . . .	*19*	*44*	*19*	*5*
Index of heterozygosis .	6·3	47·7	66·3	48·0

To estimate a general constitution of the populations, an index of heterozygosis, 100 (total of heterozygous pairs)/(total pairs involving hetero- as well as homozygous pairs), was calculated for each population. Indices are given in the table. The striking homozygosis of Siraoi population probably suggests that the present population is in process of diminution under the isolated condition. The ecological conditions seem to support this view.

KURABAYASHI, M. 1948. Differential reactivity of the chromosomes of *Paris verticillata*. *Jap. Jour. Genet.*, *23*, 22.

Patterns of differential segments, revealed by cold treatment at 0° C. for three to four days, are described for *Paris verticillata* ($2x = 10$). Every chromosome, A to E, shows differential segments which may be terminal or intercalary. Chromosomes C and D each show three different patterns: *i.e.* C_1, C_2 and C_3, and D,

D^- and D^+. As to these chromosomes, the following combinations were found in diploid and triploid plants : C_1C_2DD, $C_1C_2DD^-$, $C_1C_2DD^+$ and $C_1C_2C_3DDD$. The remaining types of chromosomes were always found to be identical or homozygous.

HAGA, T. 1946. Chromosome mutation under low temperature conditions. *Jap. Jour. Genet.*, *21*, 90-91.

In *Trillium kamtschaticum* mitosis in pollen-grains does not take place at a temperature condition of 0° C. After this finding, young uninucleate pollen-grains, in intact anthers attached to mother plants, were aged for 100 days at 0° C. Then, the aged pollen-grains were transferred from chilled to temperate condition of *ca.* 23° C. ; under this condition uninucleate pollen-grains completed their mitosis and were converted into binucleate state within one or two days.

Mitotic stage		Normal	Abnormal
Prophase	uninucleate . .	100	100
	micronuclei . .	0	0
Telophase	binucleate . .	96	8
	micronuclei . .	4	92

Nearly all the pollen-grains of three out of some twenty plants examined showed severe chromosomal and chromatidal breaks and fusions, which were comparable to the aberrations caused by very high X-ray irradiation. Prophase pollen-grains were found to be invariably uninucleate regardless of " normal " or " aberrant " plants. Whereas, nearly all the telophase pollen-grains of the aberrant plants showed numerous fragments, bridges and micronuclei. Despite these profound destructions, no case of breakage of the centromere was observed. Pollen-grains of normal plants at the same stage are nearly always binucleate, including no abnormal element (see table, a count of 100 pollen-grains in each case). This indicates that the aberrations have been induced during the resting stage which was forcibly prolonged by the ageing effect of low temperature.

SATÔ, D. 1941. A diploid plant with only one nucleolus and its bearing on the balance hypothesis of nucleolar chromosomes. *Bot. Mag.*, Tokyo, *55*, 159-163.

In *Scilla japonica* ($2n = 16, 18, 26, 34, 35, 43$) the writer (1940) found only two nucleoli at telophase in polyploid series with the following combinations of basikaryotypes, *i.e.* $18(K = B_2B_2)$, $26(K = B_1B_2B_2)$, and $34(K = B_1B_1B_2B_2)$, where K is karyotype, B_1 basikaryotype with 8 chromosomes and B_2 basikaryotype with 9 chromosomes.

Paris hexaphylla ($2n = 15$) is triploid and has one sat-chromosome and two nucleolar chromosomes, corresponding to three nucleoli at telophase. The satellite in *Paris* consists of heterochromatin and seems to be genetically inert.

Brodiæa uniflora ($2n = 12$) is diploid and has only one nucleolus, corresponding to one sat-chromosome with a long stalk. This sat-chromosome alone is attached to the surface of the nucleolus at mid- and late-prophase, though many chromosomes also are attached to this nucleolus in the early prophase. Thus it may be surmised that this sat-chromosome has so great a capacity for nucleolus organisation that the other chromosomes are made inactive in function. This discovery favours the balance hypothesis of nucleolar formation.

NAGAO, S., and MASIMA, I. 1944. Studies on the chromosomes of *Chelidonium majus*. III. Variation of karyotypes found in plants coming from different localities. *Jap. Jour. Genet.*, *20*, 59-63.

The somatic chromosome number of *Chelidonium majus* is 10 in Sapporo race instead of 12 as in English race. The Sapporo race is a translocation heterozygote,

entirely differing in its karyotype from that of English race, a normal diploid. The authors call the English race, in this paper, Karyotype I and the Sapporo race Karyotype II. Besides these the authors reported another three cases of variation in karyotypes in the materials collected from the suburbs of Sapporo, Otaru and Nagano, calling them Karyotypes III, IV and V.

HARADA, I. 1942. Chromosome studies on *Potamogeton*. *Jap. Jour. Genet.*, *18*, 92.

The somatic chromosome numbers of 21 species of *Potamogeton* were determined as follows :

	$x = 13$	$x = 14$
$2x$	*oxyphyllus* Miquel *panormitanus* Bivona-Bernardi	*cristatus* Regel et Maack *Fauriei* Miki *monoginus* Miki *Vaseyi* Robbins
$3x$	None	*apertus* Miki *kamogawaensis* Miki
$4x$	*anguillanus* Koidzumi *biwaensis* Miki *crispus* L. *dentatus* Hagström *distinctus* Bennet *Fryeri* Bennet *Maackianus* Bennet *malaianus* Miquel *natans* L. *nipponicus* Makino *perfoliatus* L. *prælongus* Wulfen	None
$6x$	*pectinatus* L.	None

These chromosome numbers can be classified into two groups : one is a polyploid series of $b_1 = 13$ ($2n = 26, 52, 78$) and the other $b_2 = 14$ ($2n = 28, 42$).

HARADA, I. 1943. Die Karyotyp der Gattung *Najas*. *Jap. Jour. Genet.*, *19*, 120-121.

Chromosome numbers of 9 species of *Najas* (submerged weeds) are reported.

N. major Allioni and *N. tenuicaulis* Miki are diœcious, and in the female plants of both species the chromosome numbers in root tip cells are $2n = 12 = 2\ l + 2\ m_1 + 2\ m_2 + 2\ m_3 + 2\ s_1 + 2\ s_2$. In the male plants of both species $2n = 13$ chromosomes are observed. One small fragment-like chromosome exists beyond the normal complement. At the metaphase of PMC-I, 6 bivalents and one such special chromosome are usually observed and approximately equal frequency of pollen-grains with $n = 6$ and $n = 7$ can be recognised. The author regards this extraordinarily small chromosome as a sex chromosome.

The following 7 species are monœcious and the chromosome dimensions are much smaller. The somatic chromosome numbers are counted as follows : *N. ancistrocarpa* Braun $2n = 12, 24$, *N. feveolata* Br. $2n = 12, 24, 34$, *N. gracillima* Magnus $2n = 12$, *N. graminea* Delile $2n = 24$, *N. minor* All. $2n = 12$, *N. oguræensis* $2n = 24$, *N. sp.* $2n = 46$.

HARADA, I. 1944. A sex chromosome of *Phyllospadix*. *Jap. Jour. Genet.*, *20*, 127-128.

Somatic chromosome numbers and occurrence of a peculiar sex chromosome were reported in *Phyllospadix* (submarine phanerogamous plants).

The female plants of *P. japonica* Makino have 20 short rod-shaped chromosomes (*ca.* 1-2 μ) in root tip cells, and the male plants have $2n = 16$ chromosomes of which 15 short rod-shaped are similar to those of the female, and one large (*ca.* 10-15 μ) V-shaped chromosome possessing one centromere. This chromosome, designated as Y, stays as a large heteropyknotic body during the resting stage. Though meiotic division in PMC was not investigated, it seems that this Y chromosome would pair with 5 short rod-shaped chromosomes.

In *P. iwatensis* Makino, the chromosome number of root tip cells is calculated as $2n = 20$.

HARADA, I. 1948. Karyotype und Entwicklungsgeschichte des fadenförmigen Pollens von Gattung *Zostera*. *Jap. Jour. Genet.*, *23*, 13-14.

Numbers and shapes of somatic chromosomes in 5 species of *Zostera* (submarine phanerogamous plants) were reported by the author as follows: *Z. asiatica* Miki, *Z. cæspitosa* M., *Z. caulescens* M. and *Z. marina* L. having respectively $2n = 12$ chromosomes and their shapes are short rods (*ca.* 1-2 μ). *Z. nana* Roth has also $2n = 12$ chromosomes whose shapes are relatively long V and J (*ca.* 2-4 μ). Six gemini chromosomes are counted in the metaphase of PMC-I of *Z. cæspitosa* and *Z. marina*.

The development of "thread-like pollen" (characteristic of some submarine weeds as *Zostera*, *Phyllospadix*, etc.) could be observed in *Z. cæspitosa* and *Z. marina*. Length of the pollen mother cells in the beginning of meiosis is 900 μ. Cell plates of the first and second divisions of meiosis develop along the long axis of the mother cells, and the pollen tetrads thus produced therefore consist of four long parallel threads. These threads separate from each other to become young "thread-like pollen" (over 1000 μ in length). The course of pollen mitosis is normal, and the mature pollen is 1700-2000 μ in length and 8 μ in width.

INARIYAMA, S. 1948. The origin of the Japanese *Lycoris* plants. *Jap. Jour. Genet.*, *20*: 87-88.

The karyotypes of 5 species of *Lycoris* known to grow in Japan and some others in China were confirmed by the author and listed in the following table:

Species	Karyotype
sanguinea Maxim.	2(11 I)
Sprengeri Comes.	2(11 I)
radiata var. *pumila* Hort.	2(11 I)
radiata Herb.	3(11 I)
straminea Lindl.	2(3 V+5 I)
aurea Herb. 14-chro. form	2(4 V+3 I)
aurea Herb. 12-chro. form	2(5 V+1 I)
aurea Herb. 13-chro. form	(4 V+3 I)+(5 V+1 I)
albiflora Koidz.	(11 I)+(5 V+1 I)
squamigera Maxim.	(11 I)+2(3 V+5 I)

(V and I stand for V- and rod-chromosome respectively)

From this table, we see that in *Lycoris* there are 4 different basikaryotypes which are represented by the formulæ 11 I, 3 V+5 I, 4 V+3 I and 5 V+1 I, respectively, and also that all the karyotypes in *Lycoris* species which the author studied consist of two or three of these basikaryotypes.

Among the five species found in Japan, *L. radiata* is a triploid, and *L. albiflora* and *L. squamigera* are both regarded as hybrids in origin from the karyological and morphological points of view. *L. radiata* carries a karyotype 33 I and its external morphological characters and habits are very similar to those of *L. radiata* var. *pùmila* with a karyotype 22 I. The chromosome behaviour in meiosis shows that this species, *radiata*, is an autotriploid. In this case the "variety" is in reality the original plant from which the "species" derived.

L. albiflora carries a karyotype (5 V+12 I), which consists of two different basikaryotypes, 5 V+1 I and 11 I. The facts that these correspond with the

basikaryotypes of *L. aurea* (5 V+1 I) and *L. radiata* var. *pùmila* (11 I), and that the external morphological characters seem to be intermediate, indicate that *L. albiflora* may be a natural hybrid raised between these two plants.

L. squamigera carries a karyotype (6 V+21 I), which consists of three basikaryotypes, two (3 V+5 I) and one (11 I), and the external morphological characters of this plant are nearly intermediate between *L. straminea* (6 V+10 I) and *L. Sprengeri* (22 I), with some inclination to the former. It seems, thus, likely that *L. squamigera* is a natural hybrid arising from the union of a diploid gamete of *L. straminea* and normal gamete of *L. Sprengeri*.

In the above three cases the supposed original or parent species are not found in Japan; all these and their derivatives grow in China. The present distribution of these supposed parent plants forces us to conclude that *L. radiata*, *L. albiflora* and *L. squamigera* must have arisen in China. Being polyploid or hybrid, they must have been more vigorous and hardier, and have had a greater power of vegetative reproduction than their parents. They thus could have reached Japan before it became separated from the Asiatic continent.

MIDUNO, T. 1943. Karyotyp einiger *Anemone*-Pflanzen. *Jap. Jour. Genet.*, *19*, 123-124.

The karyotypes of 6 species and 2 varieties of *Anemone* were observed at the mitotic divisions of root tip cells with the following results.

	2n	No. of nucleoli and of sat-chrom.	Types of chromosomes
yunnanensis	16	2	10 V, 6 I
Sieboldi	16	2	,,
Sieboldi F.	16	1	,,
altaica	16	4	,,
coronaria	16 *	5	,,
slavica	32	6	20 V, 12 I
flaccida vf.	14	2	12 V, 2 I

* 2 I chromosomes have secondary constrictions

SATÔ, D. 1947. Origin and phylogeny of karyotypes in Agavaceæ. *Jap. Jour. Genet.*, *22*, 28.

There are five different karyotypes in Agavaceæ of Hutchinson, namely, (1) *Yucca-Agave* type, (2) *Nolina* type, (3) *Phormium* type, (4) *Dracæna* type and (5) *Doryanthes* type. The phylogeny of these types was considered by the writer to be derived from the Liliaceous stock and developed to Palmæ. Accordingly the following 5 lines are postulated, based upon the karyotype analysis in these families and allies.

(1) *Eucomis* ($2n = 60$), *Hosta* ($2n = 60$)→*Yucca-Agave* type ($2n = 60$),
(2) *Ophiopogon* ($2n = 36$)→*Nolina* type ($2n = 36$)→*Trithrinax* ($2n = 36$),
(3) *Dianella* ($2n = 32$)→*Phormium* type ($2n = 32$)→*Cocos* ($2n = 32$),
(4) *Dracæna* type ($2n = 38$)→*Phœnix* ($2n = 36$), *Livistona* ($2n = 36$), *Oreodoxa* ($2n = 38$),
(5) *Doryanthes* type ($2n = 48$).

The last type has no similar karyotypes in both Liliaceæ and Palmæ, but karyotype of Lloydia ($2n = 24$) seems to be the most probable one when karyotype alteration is taken into consideration and also it is similar to Yucca-Agave type. The detailed observation was reported in *Bot. Mag. Tokyo*, *60* (1947), 31-36.

YASUI, K. 1946. On the polyploid plants of genus *Sequoia*. *Jap. Jour. Genet.*, *21*, 9-10.

Hirayosi and Nakamura determined the 2*n* chromosome number of *Sequoia sempervirens* as 66 and considered this plant as the autohexaploid plant derived from *S. gigantea* ($2n = 22$) by chromosome duplication. The author, however, holds the view that these two relic species in *Sequoia* are not closely related enough to each other for one to consider *S. gigantea* as the mother plant of *S. sempervirens*.

YAKUWA, K. 1944. On the behaviour of extra-chromosomes of *Secale cereale* L. *Jap. Jour. Genet.*, *20*, 72-73.

Crossing experiments between 14- and 16-18 chromosome ryes show the facts that in the progeny, plants with even number of extra-chromosomes can be most frequently found (table 1).

In the first metaphase of 16-chromosome rye, 8 bivalents (including supernumerary one) were found in 90 per cent. of PMC. The segregation of the bivalents is normal throughout the meiotic divisions. The observations on the division of pollen grain nucleus agree with the expectation, namely 92 per cent. of the dividing nuclei showed 8-chromosomes. Lagging of supernumerary chromosomes can be observed without exception in the anaphase.

TABLE 1

Extra-chromosome numbers of the hybrids, 14×16—18 chromosome rye

Parents	Progeny									
	0	1	2	3	4	5	6	7	8	Mean
0×2 = 1	4	...	10	...	...	...	...	...	...	1·4
2×0 = 1	3	1	5	...	1	...	...	...	...	1·5
0×3 = 1·5	...	1	3	...	...	...	...	...	...	1·75
0×4 = 2	...	1	6	...	16	1	...	...	...	3·4
4×0 = 2	...	...	3	...	2	...	...	...	...	2·8
2×2 = 2	...	...	1	2	18	...	...	...	...	3·8
4×4 = 4	...	...	...	...	...	...	1	...	1	7·0

2. PLANT BREEDING

ONO, T. 1948. Breeding of hop, with special reference to an induction of polyploids. *Seibutugaku Gyoseki*, *11*, 1-10.

By colchicine-method the author has succeeded in producing polyploids in different strains of hop, *i.e.* America, Early Zug, Shinshu-wase, Sapporo No. 6, etc.

Seedlings showing symptoms of polyploidy were carefully selected and propagated vegetatively.

The gigas plants, however, were not always pure tetraploids. Of 22 such gigas female plants, 12 produced in the progeny only triploids, 3 both diploids and triploids and the remaining 7 only normal diploids. The facts indicate that in the last 2 cases all or part of the shoots might be diploid.

Sexuality of tetraploids in hop is generally male or female and that of triploids, female or intersex. Sometimes male and female intersexes are found in both polyploids.

The triploid females are economically promising.

NOGUCHI, Y. 1943. Studies on the polyploidy in spinach. *Jap. Jour. Genet.*, *10*, 106-107.

The characteristics of polyploidy, such as large and thick leaves, large stomata small number of stomata in a definite leaf area, large flower and seed, appeared in the colchicine-induced tetraploid spinach. Delay of flowering time, about 16 days in male and 20 days in female plants, greater fresh- and dry-weight, rich water content and change of sex ratio (increase of female and bisexual plants) were also associated with polyploidy in spinach.

The most interesting and important fact from the standpoint of crop production, is the increase, about 15 per cent., in vitamin A and C content.

Date	Diploids		Tetraploids	
	20th March	15th April	20th March	15th April
Vitamin C (mg./100 g.) .	228·3	237·5	268·5	282·0
Provitamin A (mg./100 g.) .	9·8	10·8	11·3	12·3

NISHIYAMA, I. 1941. Studies on artificial polyploid plants, V. The breeding of long-fibred varieties by doubling the chromosome number in Hemp. *Bot. and Zool.*, *9*, 173-179.

C_2 and C_3 progenies of colchicine-induced tetraploid *Cannabis sativa* were compared with the original diploids and the data obtained are given in the following table.

Characters	Tetraploid (in terms of Diploid 100)
Germination of seeds	92
Stem length of seedlings	134
Habit of non-branching in young plants	195
Proportion of female plants	164
Proportion of good pollen	99
Volume of seeds	135
Weight of seeds	166
Length of fibres	147
Width of fibres	134
Strength of fibres	Over 128
Elongation of fibres	92

In general, the characters of the tetraploids exceed those of the diploids in every respect, except germination of seeds, number of good pollens and elongation of single fibre. Besides causing a remarkable increase in female plants, *ca.* 83 per cent., chromosome doubling is an excellent technique in hemp breeding as tetraploid fibres are very long and strong.

Cytological study on C_2 progenies from the tetraploid hemp has been made and their chromosome number is found to be either $4x$, $4x \pm 1$ or $4x \pm 2$ ($x = 10$) in P.M.C. In $4x$-plants, 20_{II} are usually counted although some univalents and chromosome complexes are sometimes formed. The chromosome configurations in $4x-1$ are $19_{II} + 1_{I}$ and $1_{II} + 18_{II}$ in most cases. In the latter the trivalent chromosome shows the V-shape whose two components, usually two ends, are smaller than the middle one. It is also suggested that the smaller chromosome may be X, and the larger Y, a disagreement with the works of previous authors.

NAKAMURA, M. 1942. Cytological studies in the genus *Citrus*. III. Further data on chromosome numbers. *Jour. Hort. Assoc.*, *13*, 30-40.

A chromosome counting on some hundred and fifty kinds of citrus, which represent subgenera, sections and subspecies of the genus, excluding Sect. Papeda, has proved that they were all diploid, having 9 chromosomes at the haploid phase. In addition to those, only two examples of tetraploidy were found : the Sampson Tangero and the Shikinari-mikan, a variety of *C. madurensis*. *Poncirus trifoliata* and four subspecies of *Fortunella*, including a form of *F. Hindsii*, were also determined to have 9 chromosomes in the nuclei of the gametic cells. The Temple orange, a supposed species hybrid between *C. sinensis* and *C. Tangerina*, and the Tanikawa buntan, a hybrid between *C. grandis* and another species, were found to be the diploid forms characterised by normal meiotic division. Diploid constitution and high affinity of the chromosomes were displayed by the generic hybrids, such as the Rusk citrange (*C. sinensis*×*P. trifoliata*), the Eustis limequat (*F. japonica*×*C. aurantifolia*), the Citradia (*P. trifoliata*×*C. Aurantium*) and Thomasville citrangequat (*F. margarita*×*C. sinensis*×*P. trifoliata*).

NISHIYAMA, I., and TAKASUGI, K. 1949. Studies on artificial polyploid plants, XII. Investigations on the growth and yield of an autotetraploid radish, Minoyonbai-daikon. *Research of Food-Science*, Kyoto Univ. No. 1, pp. 1-10.

Comparative studies on the growth of two diploids (Minowase and Miyashige-daikon, $2n = 18$) and an artificially induced tetraploid radish (Minoyonbai, $2n = 36$) were made by weekly measuring of plant height, number of leaves, length and diameter of root taps, etc., of about 50 individuals in each category, for two successive seasons. The results obtained clearly showed that Minoyonbai ($4x$) and Minowase ($2x$) are superior to Miyashige ($2x$). The growth curves of the former two showed a higher vigour in Minoyonbai during the seedling stage and at the end of the season, but not in the middle of the season. Minowase is a well-known early variety in Japan.

" Pithy (porous) tissue " was found in Minowase to reach 75 per cent. in 90 days, in Miyashige 40 per cent. and in Minoyonbai only 20 per cent.

In different climatic regions in Japan during 1944-1947 the yield of Minoyonbai was consistently higher than that of the diploid varieties. Minoyonbai is probably suitable for cultivation in summer as it does not bolt in this season.

MIZUSHIMA, U. 1944. Studies on some auto- and allopolyploids made in *Brassica*, *Sinapis*, *Eruca* and *Raphanus*. *Agric. and Hort.*, *19*, 743-744.

In spite of the formation of varying numbers of quadrivalents, meiosis in the autotetraploids obtained from 8(B)-, 9(C)-, 10(A)-chromosome *Brassica* species, *S. arvensis* (S), *S. alba*, *E. sativa* and *R. sativus* (R) was carried out fairly regularly resulting, in most cases, in the formation of gametes with $2x$ number of chromosomes, whereas in those obtained from the digenomic species, such as *B. carinata* (BC), *B. juncea* (AB), and *napus* (AC), the division went on quite irregularly, showing little possibility for the formation of gametes with $2x$ number. From these facts the author has concluded that, as far as the materials are concerned, in breeding desirable crops by means of allopolyploidy, only $4x \times 4x$ crosses are applicable among the so-called monogenomic species, as for the digenomic ones such crosses will result only in aneuploids.

In the 3 digenomic tetraploids made by the author, AB, AS and AR, the meiosis was quite regular, forming exclusively the corresponding number of bivalents at MI. While in the 4 digenomic hexaploids AAB, AAC, BBC and ARR, as well as in the 2 trigenomic hexaploids, ABC and BCS, there appeared frequently, besides some quadrivalents, varying numbers of univalents which made the division considerably irregular. The quadrivalent formation in the 2 trigenomic hexaploids was concluded presumably to be due to the partial homology among the 3 different genomes coexisting. All of the allopolyploids, except ABC and BBC, were plants

of the first generation just having reached their flowering stage and their fertility was unknown.

MIZUSHIMA, U. 1944. Some amphidiploids in Cruciferæ. I-IV. *Breeding and Hort.*, *2*, 401-404, 441-444, 481-483, 515-516.

Seven amphidiploids obtained through colchicine and acenaphthene were observed to be intermediate between their respective parents and showed clear gigas nature against untreated sister F_1 hybrids. Their origin, gametic chromosome complements and numbers are given in table 1.

TABLE 1

B. carinata (BC, $n = 17$)	× *B. campestris* (A, $n = 10$)	AABBCC, $n = 27$
B. carinata	× *B. nigra* (B, $n = 9$)	BBBBCC, $n = 25$?
B. napus (AC, $n = 19$)	× *B. pekinensis* (A, $n = 10$)	AAAACC, $n = 29$
B. rapa (A, $n = 10$)	× *R. sativus* (R, $n = 9$)	AARR, $n = 19$
R. sativus	× *B. rapa*	AARRRR, $n = 28$
B. campestris	× *S. arvensis* (S, $n = 9$)	
B. rapa	× *S. arvensis* (S, $n = 9$)	AASS, $n = 19$
B. carinata	× *S. arvensis* (S, $n = 9$)	BBCCSS, $n = 26$

(AARRRR was obtained from an occasional triploid hybrid)

In selfing, the untreated hybrids were utterly sterile, whereas the amphidiploids showed varying degrees of fertility ranging from 60-10 per cent. The MI configuration in the microsporogenesis in the untreated hybrids and the corresponding amphidiploids are given in table 2.

TABLE 2

Untreated hybrids	Amphidiploids	Self-fertility
ABC $(9\text{-}1)_{II}+(9\text{-}25)_{I}$	$(7\text{-}0)_{IV}+(13\text{-}27)_{II}$	*ca.* 50 per cent.
BBC $8_{II}+9_{I}$	$(8\text{-}1)_{IV}+(9\text{-}23)_{II}$	60 ,,
AAC $10_{II}+9_{I}$	$(9\text{-}3)_{IV}+(11\text{-}23)_{II}$	10 ,,
AR $(0\text{-}5)_{II}+(19\text{-}9)_{I}$	19_{II}	...
ARR $9_{II}+10_{I}$	$(1\text{-}9)_{IV}+(26\text{-}10)_{II}$	...
AS $(0\text{-}5)_{II}+(19\text{-}9)_{I}$	19_{II}	50 ,,
BCS $(0\text{-}8)_{II}+(26\text{-}1)_{I}$	$(4\text{-}0)_{IV}+(18\text{-}26)_{II}$	60 ,,

(AARR and AARRRR were just in the flowering stage)

HOSODA, T. 1946. Fertility of colchicine-induced amphidiploids between *Brassica* and *Raphanus*. *Agric. and Hort. Tokyo*, *21*, 515.

F_1 seeds from the crosses *Brassica rapa* × *Raphanus sativus* were treated by colchicine and from them 4 allotetraploids and 1 allohexaploid were obtained.

The number of seeds resulted from these plants was unexpectedly small, as shown in the following table, compared with that of pods. As some other allotetraploids or allohexaploids obtained by colchicine treatment from *Brassica* species hybrids were reported to show better fertility, it may be difficult to conclude that the high degree of sterility of the author's *Brassico-Raphanus* and *Raphano-Brassica* hybrids was simply caused by the after-effect of colchicine.

Fertility of Brassico-Raphanus *and* Raphano-Brassica F_1-*s*

Parents	Amphipolyploids (F_1)	Nos. of pods	Nos. of seeds
Turnip × Radish	Tetraploid	49	5
,, × ,,	,,	38	1
,, × ,,	,,	42	0
Radish × Turnip	,,	45	23
,, × ,,	Hexaploid	16	19

HOSODA, T. 1946. On the dimension of F_1 seeds in crosses among *Brassica*, *Sinapis* and *Raphanus*. *Agric. and Hort. Tokyo*, *21*, 516.

A comparison of the dimensions of parental and F_1 seeds obtained from crosses among *Brassica*, *Sinapis* and *Raphanus* was made and it was ascertained that true hybrid seeds are smaller than false hybrid seeds. The hybridity of F_1 seeds was justified by the observation of the morphological characters of the seedlings.

KONDO, N. 1942. A new *Raphanobrassica* from the cross *4x-Raphanus sativus* L. × *4x-Brassica oleracea* L. *Jap. Jour. Genet.*, *18*, 126-130.

A new *Raphanobrassica* was obtained from the cross between autotetraploid *Raphanus sativus* L. and *Brassica oleracea* L. The parents were offsprings of autotetraploids induced by colchicine treatment.

At the first metaphase of PMC, 1-7 polyvalents and 0-1 univalent were formed in the autotetraploid *Raphanus*. In the autotetraploid *Brassica* 2-5 polyvalents and 0-2 univalents were observed.

There is indication of incompatibility in some cross-combinations as shown in the following table (plants 1 and 2) :—

Crosses	No. of flowers pollinated	No. of seeds obtained
R. sativus (4*x*)-1 × *B. oleracea* (4*x*)-1	19	20
R. sativus (4*x*)-1 × *B. oleracea* (4*x*)-2	24	22
R. sativus (4*x*)-2 × *B. oleracea* (4*x*)-1	10	0
R. sativus (4*x*)-2 × *B. oleracea* (4*x*)-2	12	0

(The reciprocal crosses were unsuccessful)

Out of the 35 seeds obtained, 29 gave rise to mature plants which were morphologically intermediate between their respective parents. Cytological investigations of the 24 individuals which bore flowers revealed that 24 plants had 36 somatic chromosomes and the chromosome configuration at meiosis was 18_{II}. One was hypertetraploid ($18_{II}+1_I$).

KIHARA, H., und YAMADA, I. 1942. Ein Fall Plasmonwirkung auf das artfremde Genom. *Seiken Ziho*, *1*, 38-45.

The present studies deal with the problems of genome and plasma in interspecific hybrids of diploid *Aegilops*, viz. *Ae. comosa* × *Ae. uniaristata*, the genome constitutions of which were established by the senior author (Kihara, 1937) as M and M^u respectively.

F_1 plants have 0·92-0·96 per cent. of good pollen and 0·82-1·1 per cent. grain set. The fertility agrees with the theoretical percentage, expected from the mode of chromosome behaviour during maturation divisions, where it is assumed that one chromosome is exchangeable between the semi-homologous M- and M^u-genome.

All the plants from the back-crosses, F_1 ♀ × *Ae. comosa* ($MM^u \times MM$) were fertile and had the same morphological characters as *Ae. comosa* (MM), and those from F_1 ♀ × *Ae. uniaristata* ($MM^u \times M^uM^u$) were sterile and had the same morphological characters as F_1 (MM^u).

From the data obtained, it can be said that in F_1 (MM^u) plants with comosa plasma, only one type of gamete with perfect M^u (*Ae. comosa*) genome is functional, gamete with M^u (*Ae. uniaristata*) genome is lethal.

KIHARA, H. 1944. Chromosome conjugation in a rose hybrid, *Rosa multiflora* × *R. rugosa*. *Jap. Jour. Genet.*, *20*, 55-58.

Cytological studies on a species hybrid between two diploid species (*R. multiflora* ♀ × *rugosa* ♂) were carried out. The hybrid is found to have 1-7 (mostly 5-7)

pairs at first metaphase and the bivalents exhibit normal chromosome behaviour throughout the course of the maturation divisions. The univalents split longitudinally in the first anaphase and they are distributed to the poles at random in the second anaphase.

The hybrid produced only empty pollen grains and artificial pollination with *multiflora*-pollen gave no fruit. The sterility seems to be caused by physiological conditions affecting growth of the micro- and macrospores during the gametogenesis.

KIHARA, H., and NISHIYAMA, I. 1947. An application of the sterility of autotetraploids to the breeding of seedless water-melons. *Seiken Zihô*, *3*, 93-103.

Among approximately 500 seedlings of water-melons treated with colchicine in the fall of 1942, many were found to have an increase in size in stomata, pollen grains and seeds. From the seeds obtained from open pollination from these individuals, tetraploids and triploids were found. The appearance of triploids was due probably to free pollination of induced C_1 tetraploids with diploid pollen.

Cytological studies showed that in the tetraploids II_{IV} can be counted, yet sometimes their 4 elements separate into 2 bivalents. In the triploids, II_{III} are formed in I-M, but failure of pairings such as $I_{III}+I_{II}+I_I$ were also noticed.

Giant nuclei, irregular in shape, due perhaps to failure or incompleteness of meiotic divisions, and pollen grains connecting with each other as in tetrad, were occasionally observed.

Pollen sterility of the triploid was high, 58-74 per cent., that of the diploid and tetraploid was 1-4 per cent. Triploid individuals appear to be most vigorous, next come the diploids; least vigorous of all are the tetraploids. Triploid and tetraploid plants set normal fruits. Though the former have practically no seed, cases in which a few seeds whose seed coats have more or less normal appearances were found in triploid fruits.

In the production of triploids, no viable seed could be obtained when *2x* plants were used as females.

3. ANIMAL CHROMOSOMES

OGUMA, K. 1946. Karyotype and phylogeny of mantis. *La Kromosomo*, Vol. 1, *1*, 1-5.

After chromosomal survey of 15 species of Mantidæ, collected by Dr J. J. Asana in East India and by the author in Japan and adjacent territories, it is proposed that the family Mantidæ should be divided into two sections, Monosomata and Trisomata, in respect to the constitution of sex-chromosomes.

MONOSOMATA.—The most subfamilies studied, viz. *Eremiaphilinæ*, *Scizocephalinæ*, *Hymenopodidæ*, *Toxoderidæ*, *Vatinæ*, and *Empusinæ*, are to be classified into this section. The chromosome number in the male varies from 15 (*Didymicorypha lanceolata*) to 35 (*Humbertilia indica*) and all chromosomes are V-shaped in varied size. The sex-chromosome is V-shaped, of enormous size, and never accompanies the Y. This karyotype seems to be more primitive as compared with that of the next section.

TRISOMATA.—Species included in subfamily *Mantinæ* are to be classified in this section. The chromosome number in males is 27 in every species and the shape of chromosomes is V as in the preceding section. But the sex-chromosomes are constituted of three distinct ones, X_1, X_2 and Y. These two X's seem to have been derived from a single huge-sized X-chromosome through a certain mechanism as demonstrated by White (1940) for instance. This fact suggests to us therefore that Trisomata should be an advanced group in phylogeny.

Further, in three species of *Hierodula*, size diminution of sex-chromosomes occurs in two directions, first in X_2 (*H. ventralis*), secondly in Y (*H. tenuidentata*). From such a phenomenon it will be expected that in some unknown species the Y-chromosome may entirely disappear, only two X's being left behind. But species, if actually present, should be found in Trisomata. In this respect, therefore, the information of two X's in *Empusa* by Erazi (1940) is quite doubtful.

YAMASHINA, Y., and MAKINO, S. 1946. A study on the chromosomes of pigeons and doves. *Siebutu*, *1*, 92-100.

The chromosomes of three species of the Columbidæ were studied in the germ cells from embryonal gonads and adult testis. The chromosome numbers obtained are shown in the following table :—

Species	*2n*	*n*	Sex-chromo.
Columba livia var. *dom.* . . .	80 spg, 79 oog.	40 ♂ (I)	XO in ♀
Streptopelia decaocto var. *risoria* . .	76 spg, 75 oog.	38 ♂ (I)	,,
Streptopelia orientalis	75 oog.	...	,,

The male *2n* complex of the pigeon consists of five pairs of V-shaped elements together with the remaining rod-shaped ones, while in the female the fourth element of the medium V-shape is destitute of its mates. On this basis the fourth V-element is the sex-chromosome.

The chromosome complements of two species of doves are similar. In each of them the male complement contains 7 pairs of V-elements and the remaining pairs are telomitic rod-type of varying sizes. The fourth largest V-element is single in the female cell, and therefore this element can be accepted as the sex-chromosome.

The F_1 between *S. decaocto* ♀ and *S. orientalis* ♂ had apparently normal genital organs, in the male and in the female. The male meiotic behaviour of chromosomes seems to be regular in showing 38 bivalents.

YASUZUMI, G., KITA, T., and OKAZAKI, A. 1948. On the molecular structure of salivary gland chromosomes. II. *Jap. Jour. Genet.*, *23*, 59-60.

The authors' histochemical and electron microscopic investigation on salivary gland chromosomes of *Drosophila virilis* has revealed the chemical constituents of the chromosomes as follows :

1. The euchromatin region contains amino acids, that is, tyrosine, tryptophane, arginine and others, and also enzymes, namely SH-group, Dopa's oxidase, peroxidase, and others.

2. Histidin is found as a side chain of chromonema.

3. In the matrix, glycogen is found with the help of Best's carmin method, diastase digestion, iodine, Molish's and Bauer's reaction.

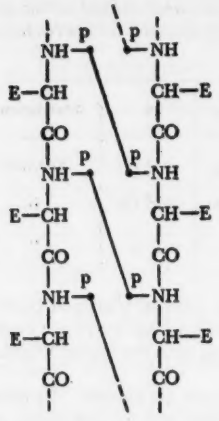
4. The isoelectric point of chromosomes was determined by the dyestuff absorption methods : euchromatin, *p*H 3·2 ; heterochromatin, *p*H 1·35-1·90 ; chromonema, *p*H 7·65.

5. After treatment with spleen nuclease, the chromosomes are no longer stained with the Feulgen's techniques. The value of an isoelectric point of the thus treated euchromatin parts is at *p*H 9·8.

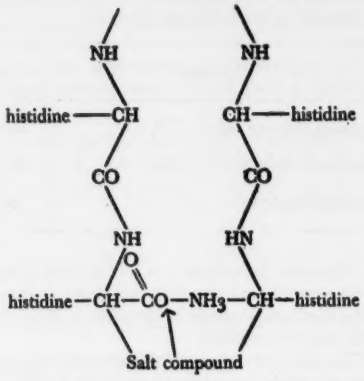
6. A new method devised by the authors for arginine reaction (*p*-dichloramidobenzoic acid dissolved in normal sodium hydroxide solution) has yielded very good results. Through this method euchromatin clearly gives a positive reaction while heterochromatin is negative, and tryptophane reaction produces the same result. Thus the difference in chemical composition between the euchromatin and heterochromatin has been confirmed.

7. The kind of basic protein of the salivary gland chromosomes was decided by staining with brillantazurin B as well as isoelectric point to prove that the greater part of the basic protein was histone, a further part of it being a non-histone protein.

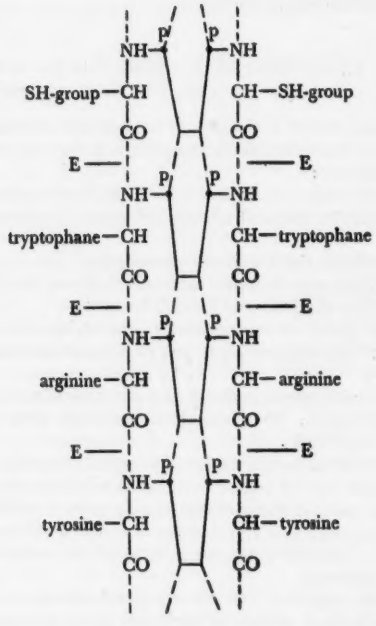
8. The protein of chromosomes is probably formed by Fischer's polypeptide link, and has various kinds of amino acids as its side chains.



Schematic molecular structure of the heterochromatin (β-polypeptide)



Schematic molecular structure of the interband (α-polypeptide)



Schematic molecular structure of the euchromatin (β-polypeptide)

MAKINO, S., and MOMMA, E. 1947. A new smear acetocarmine method for the demonstration of spiral structure of chromosomes in the grasshopper. *Kagaku, 18,* 36-37.

The experiment was carried out mostly with male germ cells of Podisma (Acrididæ) as material.

Pretreatment.

(1) With cold water. The testes of the grasshoppers are submerged without squashing either in tap or distilled water in a glass tube for 20-30 minutes at room temperature, or for 60 minutes at 12-13° C.

(2) With hot water. Glass tube containing distilled water and fresh testes are immersed in hot water kept at about 50° for 20 minutes.

(3) With ammonia solution. The testes are submerged *in toto* in 0·01 per cent. ammonia solution and kept at 12-13° C. for 40-45 minutes.

Staining. The pretreated testes are squashed on a slide and the cells, gently smeared, are covered with acetocarmine. To secure even penetration, leave it 2-3 minutes before placing cover-slip in position.

Amongst the three procedures attempted, the first one, especially when using cold water at 12-13° C. produces, as a whole, uniform and constant results. When the time of pretreatment has been suitable, the spiral structure of the chromosomes is observable in almost all of the dividing cells. One of the advantageous points of this technique is that in the material prepared, both the cells and the chromosomes are strikingly prevented from shrinking.

4. ANIMAL AND HUMAN BREEDING

NOZAWA, K. 1943. Genetics of Thecamœba. I. Heritable shell abnormalities found in *Arcella hemisphærica* and *Centropyxis aculeata. Jap. Jour. Genet., 19,* 189-199.

Two kinds of shell abnormalities, double-lobed and slitted, were obtained in old cultures of both these species. The phenotypes are similar to those previously described by Reynolds (1923) and Jollos (1924) in *Arcella polypora.* They were inherited only by a few of their progeny, most of which were quite normal individuals.

It has been shown that the time (2 to 12 months) during which the progenitor of the clones had been kept in old culture favouring the appearance of abnormal individuals, had nothing to do with the degree of inheritance of the characters in question. Though old cultures are liable to yield more shell abnormalities than new ones, as found by the previous authors, it is somewhat doubtful whether the change is a dauer-modification in Jollos' sense (1924, 1939). In dauer-modifications we must make a distinction between the mechanism of appearance of the new character and of its inheritance.

There is no causal relation between the abnormality of the shell and the nuclear number in *Arcella hemisphærica.* (where the nuclear number is heritable). The shell of the trinucleate line is larger than that of the binucleate line, the ratio being 1·24 to 1·00 in diameter.

INABA, F. 1944. Genetical studies on *Habrobracon pectinophoræ.* I. Sex-linked inheritance and sex-determination. *Jap. Jour. Genet., 20,* 27-47.

The recessive mutant body-colour character, ebony (eb) seems to be sex-linked. The recombination value of ebony and the sex-gene is 19·73±0·79 per cent. The back-crosses of heterozygous females with ebony males segregate into three types, namely, deficiency, excess and sub-equality, according to whether too few ebony daughters or too many ebony daughters or nearly as many ebony daughters as wild-type daughters are given.

When heterozygous daughters of a single ebony male are divided into two groups and one group is mated with their wild-type brothers and the other group

with unrelated wild-type males, the former gives either excess (75·00±2·18 per cent.) or deficiency type (15·28±4·72 per cent.) heterozygous daughters, while the latter invariably gives equality-type heterozygous daughters with a slight inclination towards deficiency (46·71±2·46 per cent.) or excess (55·64±2·54 per cent.). Thus sex-linkage of ebony disappears in the out-crosses, while it is clear in the close-crosses.

Back-crosses of F_1 heterozygous females of the out-crosses by their own ebony sons yield daughters among which ebony is either deficient (13·37±1·49 per cent.) or in excess (82·43±4·64 per cent.). The fact that the sex-linkage of ebony distinctly reappears with inbreeding, even directly after out-crossing, favours Whiting's multiple allele theory more than Snell's multiple factor theory of sex determination in *Habrobracon*.

A new theory of compound sex-gene is proposed by the author, namely, the male-determining gene occupies a segment of the X-chromosome and consists of subgenes like *xa*, *xb*, *xc*, . . . etc., which are characteristic to each strain and partially homologous with one another. Ebony or fused links with one of these subgenes. Female-determining genes are scattered on the autosomes, which are of different strength and are linked with different mutant genes. In haploids, the male-determining genes dominate over female-determining genes and produce males. In diploids, when the recessive male-determining subgenes become homozygous or when two different subgenes have large common homologous parts, these genes dominate over autosomal female-determining genes and produce males. Any heterozygote of male-determining subgenes, without any homologous parts in common, produces a female, since the male-determining gene is dominated over by the female-determining gene.

OMURA, S. 1948. Selective fertilisation in *Bombyx mori* (3). *Jap. Jour. Genet.*, *23*, 40-41.

Two mutants for the colour of youngest larval skin, " dominant chocolate " and " black " were used. They were of different strains, the former being a pure strain continued 6 generations by sib matings, while the latter was a hybrid of economic races. The males of the latter had been known to be much stronger in fertilising ability. In double matings more than 90 per cent. of the larvæ hatched out from every batch were " black," regardless of the order of mating.

On the " black " males, the following treatments were done : (1) Removing one of the testes at the 2nd day of the 4th larval stage ; sperms may be reduced to some half in quantity and may be under good nutritional condition. (2) Bleeding at full matured larval stage ; sperms may be reduced in quantity, being under poor nutritional condition. (3) Cooling at 5° C. for 2 weeks at the middle stage of pupa ; sperms may be affected in quality and quantity. (4) Cooling at 5° C. for 2 weeks at adult stage ; sperms have been known to age, though the quantity may not be reduced.

Double matings were made by any of these treated males of " black " and untreated males of " dominant chocolate." In all cases selective fertilisation occurred clearly, every batch being occupied mainly by " black " larvæ. The results may be interpreted to show that the difference in the strength of the fertilising ability of the male moth is not due to the quantity but to the quality of the sperms.

NISHINA, Y., and MORIWAKI, D. 1941. An experiment on the grouping of gene mutations in *Drosophila melanogaster*. *Jap. Jour. Genet.*, *17*, 171-174.

The object of this experiment was to see whether X-rays also give rise to simultaneous occurrence of gene mutations in one male as well as in one chromosome. The ordinary " CIB " method for sex-linked lethals was used and the F_2 maleless culture, which is regarded as the result of the presence of a lethal or lethals located in the treated X-chromosome, was carried to the next F_3 offspring. It is possible

to determine approximately whether the lethal mutation is either one in one chromosome or many in one chromosome, although by this method the determination is possible only when two or more lethals are situated far enough apart from one another. In this way, out of 17 F_2 maleless cultures induced by X-rays, there was no case with two or more lethals per chromosome, whereas in our previous work in which neutrons were used, out of 18 F_2 maleless cultures tested there were 8 samples with 2 or more gene mutations in the same chromosome.

The authors have further found that " grouping in one male " could be raised with X-rays and that an extremely high rate of " grouping in one chromosome " has been observed.

YAMASHINA, Y. 1943. Study on sterility in hybrid birds. IV. Cytological researches on hybrids in the family Phasianidæ. *Jour. Fac. Sci. Hokkaido Univ.*, Ser. VI, Zool. *8*, 307-386.

The chromosome number in several breeds of domestic fowl is defined, the diploid number of the males being 78 and that of the females 77. The diploid chromosomes of two subspecies of common pheasant (*Phasianus colchicus karpowi* and *P. c. versicolor*) were found to be 82 in the male and 81 in the female, while the haploid number in the primary spermatocyte is 41 (male, *n*) without exception. Many F_1 hybrids were produced from various combinations of these parents and were found to possess the chromosome complex of the two parents, 80 in the male hybrid and 79 in the female. The male germ cells of this hybrid do not advance beyond the pachytene stage of the primary spermatocyte and thereafter degenerate, failing to form the metaphase spindle of the first division. Degeneration of the oocyte seems to take place during the leptotene stage. Thus, complete sterility of this hybrid is proved cytologically.

The chromosome numbers in the males of the Japanese copper pheasant, *Syrmaticus soemmerringii scintillans*, the common pheasant, *Phasianus colchicus*, and the golden pheasant, *Chrysolophus pictus*, are identical, 82 in diploid and 41 in haploid. The morphological characteristics differ only in the length of the *b*-chromosome, which is longest in *Syrmaticus* and shortest in *Chrysolophus*.

In the testes of the hybrid between the copper pheasant and the golden pheasant the majority of the germ cells form the metaphase spindle of the first meiotic division, while some degenerate and fail to complete the second meiotic division. A few pass through the second division, most of them forming abnormally-shaped spermatozoa. Only a few develop into normal spermatozoa, thus indicating the partial fertility of the male.

In the male hybrid between the Lady Amherst pheasant, *Chrysolophus amherstiæ*, and the golden pheasant, *C. pictus*, no abnormality is found. The chromosomes pass through two meiotic divisions normally and develop into functional spermatozoa, so the male hybrid is completely fertile.

Sterility in the hybrids *Gallus* × *Phasianus* and *Syrmaticus* × *Chrysolophus* is of the chromosomal type. Another factor leading to sterility is the antagonistic relationship between the nuclear element coming from the male parent and the cytoplasm of the female parent. In cases where the chromosomal dissimilarity between the parent species is very large and the nuclear elements are not harmonious, the degeneration of the hybrid germ cells occurs very early in meiosis, as in *Gallus* × *Phasianus*. If the chromosomal dissimilarity between the parents is small and the nuclear cytoplasmic relation is quite harmonious, the degeneration of the hybrid germ cells takes place in the latter stage of meiosis, as in *Syrmaticus* × *Chrysolophus*.

KOMAI, T. 1946. On the inheritance of black, yellow and tortoiseshell colour in cats, with special reference to the problem of the tortoiseshell male. *Seibutu*, *1*, 1-7.

The yellow is a sex-linked gene, while black is autosomal. This may be inferred not only from breeding records but also from the census of a cat population. Yellow

colour is commoner in males than in females, while such disparity is not found among black cats. The tortoiseshell male apparently results from duplication of colour and sex gene sets on an X-chromosome by unequal crossing over which takes place between the X's of a tortoiseshell female. Existing records of the appearance of such males as well as similar puzzling breeding records can be accounted for by this theory.

KOMAI, T. 1946. Two hereditary characters of the human tongue. *Jap. Jour. Genet.*, *21*, 10-11.

The ability and inability to roll the lateral edges of the tongue is due to a single set of Mendelian allelic genes. The gene for the ability is dominant over the gene for the inability, but the penetrance is somewhat incomplete especially in men. In some people the frenulum linguæ extends forward nearly to the tip of the tongue, and hinders the movement of the tongue, especially in pronouncing the *l* or *r* sound. Three pedigrees showing dominant inheritance of this abnormality are presented.

The incidence of the people devoid of the ability to roll the lateral edges of the tongue varies considerably with age ; it amounts to nearly 50 per cent. among 6-year old children, and decreases gradually with the advance of age until 12 when it is 26 per cent. among males and 18 per cent. among females. It is to be noticed that these figures, obtained for nearly 7000 Japanese, show a significant difference from the corresponding figures reported for Americans—34 per cent. among men and 28 per cent. among women (Urbanowski and Wilson, 1947, *Jour. Hered.*, *38*, 365-366), and also from those reported for Chinese—41 per cent. among men and 30·6 per cent. among women (Liu and Hsu, 1949, *Jour. Hered.*, *40*, 18-21).

REVIEWS

THE THEORY OF INBREEDING. By R. A. Fisher, 1949. Edinburgh : Oliver and Boyd. Pp. viii+120. 10s. 6d.

The quantitative study of inbreeding began, as Professor Fisher points out, with Darwin. He showed by careful experiment that inbreeding often has effects, such as loss of vigour, likely to incur a selective disadvantage, but that these do not indicate permanent injury to the stock, for vigour is fully restored in the first generation bred by crossing inbred lines. Darwin was concerned to discover the advantage which devices encouraging cross fertilisation give to their possessors rather than to elucidate the cause of the inbreeding depression itself. Indeed no effective approach to this latter problem was possible until Mendel's rules of inheritance were discovered and applied. In general terms, it has become apparent that, genetically speaking, the effect of inbreeding is to increase the frequency of homozygotes of all kinds in the inbred family or population, and that, as a result of dominance, the average expression in a series of homozygotes showing the same gene frequencies as the heterozygotes from which they came, will not be the same as the expression in those heterozygotes.

In practice the use of inbreeding has proved to be of immense importance in crop production. Of the maize grown to-day in the U.S.A., some 90 per cent. is produced by the deliberate crossing of inbred lines—lines which, because of their near homozygosity are genetically reliable and phenotypically stable. They can be tested for combining ability year after year with the confidence that the results of these tests may be legitimately pooled for the detection of fine differences, and that once a good performance has been established it can be repeated at any time, on any scale, for the purposes of commercial production. Similar advantages are being obtained in other crops, sugar beet and onions for example, as the technical problems of inbreeding and crossing are overcome. They are also being sought in domestic animals, though here the special difficulties of slow breeding and expense in maintenance must make progress less speedy.

Three distinguishable, though inter-related, problems present themselves therefore to the geneticist who is interested in the application of his science to crop and stock improvement :—the problem of discovering how, in a quantitative sense, the genotype will respond to the many various modes of inbreeding that are possible ; the problem of how an inbreeding programme can be most efficiently designed for the achievement of its purpose ; and the problem of how the more valuable products of inbreeding can be most efficiently separated from the less valuable for use in the production of commercial crops and stocks.

As the name of the book suggests, Professor Fisher is not specifically concerned with the third problem in *The Theory of Inbreeding*, though we may note that his own methods of conducting field trials have been of major importance in the testing of maize inbreds in the U.S.A., and that his Appendix C on the practical function of inbreeding must be of interest

to all plant and animal breeders. He addresses himself particularly to the question of the genetical achievement to be expected from inbreeding programmes of diverse kinds in species with various genetical constitutions and breeding habits. After an introductory chapter on the history, theory and use of inbreeding, the matter of segregating inbred lines is taken up. Their uses are discussed and the practical problems, numbers of individuals required in a generation, expectation of a suitable mating, and the complications introduced by linked genes, are analysed in special relation to mice. The next chapter is devoted to the development of a comprehensive approach to the mathematical problems which arise in working out the consequences of a system of mating. Initially this is developed in relation to consistent sib-mating, but it is later applied to other systems of mating, to the evaluation of the effect of irregularity in the mating system, to the effects of enforced heterozygosity of gene differences (including the sex difference), and to the inbreeding of species showing polysomic inheritance. Of the three appendices, one has already been mentioned. Another deals with the special problems of efficiency and time which arise when inbreeding a species bearing a single offspring at a birth, and the remaining one discusses the efficiency of self-incompatibility mechanisms, including tristyly, in diminishing the frequency of mating between near relatives in plants.

It will be clear from this list of contents that the book is most comprehensive in its coverage of the analytical problems which arise both in foreseeing the consequences of an inbreeding programme and in designing programmes to fit the special requirements of particular species or aims. This coverage is achieved within a relatively small compass by the generality of the methods developed. Series showing the rise of homozygosis to be expected under particular mating systems and particular circumstances have long been familiar in the genetical literature : indeed the first, that for continued selfing, was given by Mendel himself. Fisher's approach contrasts with these in that it can be applied to any system of mating in any circumstances, and permits irregularities to be taken into account. It is made all the more valuable in this latter respect, by the unexpected effects which irregularities may have. Sib-mating and parent-offspring mating are, when carried out in separate programmes, equally efficacious systems of inbreeding. Yet either, if occurring as an irregularity in one generation of a programme based on the other, results in a loss of efficiency of about 15 per cent. in terms of generation time. This result also serves to illustrate the essentially practical value of many of the conclusions reached ; for it is upon such a consideration that the breeder carrying out an inbreeding programme must base his judgment as to whether a readily available but irregular mating would be more expeditious than the continuation of the regular system by a mating which may take some time to mature.

A further result which brings out the great value of the method in the consideration of special cases is the demonstration that, in spite of the loss of efficiency caused in the general case by the intercalation of sib-mating in a series of parent-offspring matings and *vice versa*, systems in which matings of these kinds are deliberately included in a cycle can be considerably more efficient than the uniform systems themselves in rendering homozygous marked sections of chromosomes, such as the differential segment of the X. Again the practical aspect is clear for, in deciding whether

to use such a cyclical system, the inbreeder must compare the increase of efficiency achieved for the marked segment with the reduction in efficiency for the remainder of the chromosomes. Fisher shows how this can be done with precision.

Enough has been said to show how valuable a book *The Theory of Inbreeding* is. Geneticists may complain that it is not easy reading. Yet to those possessed of persistence and a little mathematical accomplishment, the difficulties will smooth themselves out. Indeed, chapter III is an introduction to matrix algebra, and its genetical uses, which at least some of us would have found useful on many occasions in the past. And whatever trouble the reader must take in following the mathematical methods used, he will be amply repaid not only by the insight he gains into the ramifications of inbreeding but also by the increased confidence and precision which he can bring to the practical problems of designing and carrying out inbreeding programmes, whatever his aim in their use and whatever the vagaries of the species with which he may be concerned. K. MATHER.

THE BIOLOGY OF MENTAL DEFECT. By Lionel S. Penrose. Sidgwick and Jackson, 1949. Pp. xiv+285. 21s. Preface by Professor J. B. S. Haldane.

The perennial problem of mental deficiency may be approached from the point of view of a variety of special studies, such as medicine, genetics, psychiatry, law or sociology. From any one of these angles an exhaustive treatment could be undertaken. The successes of specialists in their own fields have sometimes given rise to an unjustifiable optimism about the final solution of the problem. Early and accurate diagnosis, hormone therapy, shock treatment, psychological training, legal protection for both the defective and the society he lives in, and positive and negative eugenics, all have their contributions to make. A synoptic survey of the whole field shows that the problem is much more complex than was originally suspected, but the ultimate alleviation, cure and prevention of mental deficiency on a considerable scale will no doubt be achieved by a synthesis of many different methods and researches.

In his earlier book, *Mental Defect*, Professor Penrose described the subject as providing great opportunities for research, especially in the fields of genetics, medicine and sociology. Advances made in the fifteen years which have elapsed since then now warrant a much more integrated treatment. The ramifications of the subject are enormous, and " to limit the task here, medical and psychopathological conditions encountered in mental deficiency practice are discussed chiefly within a framework of genetics ". The aim of the book is to present the problem of mental deficiency from the broader aspect of human biology so that its study can be more readily related to the remainder of sociology and medicine.

Penrose commences with a general historical survey and then goes on to consider the incidence, definition and measurement of mental deficiency. This is followed by a chapter on the principles of classification. Next, there is a discussion of the rival claims of nature and nurture to be the causative agents of mental defect. The subsequent five chapters are especially concerned with the genetical aspects of the problem, dealing with dominant, recessive and sex-linked defects ; the genetics of intelligence and its relation to differential fertility ; and methods of analysis in human

genetics. There is a chapter devoted to mental disorders, as distinct from mental defects, and a final chapter on treatment.

In treating so vast a subject as mental deficiency it is of course impossible to give full weight to every aspect and a considerable selection of topics for discussion is inevitable. However, there are a few notable omissions which should be mentioned. For example, the section on " Linkage " in the chapter entitled " Methods of Analysis in Human Genetics " makes no mention of the important method of u-statistics first developed by R. A. Fisher in 1935 and later greatly extended by D. J. Finney from 1940 onwards. Appendix 9 does give Penrose's sib-pair test but this is less efficient than using the appropriate u-statistic. Again, in the same chapter, the section on " Mendelian Ratios " fails to emphasise the importance of the method of ascertainment, and does not even explicitly mention and distinguish the Proband and Sib methods.

Another criticism which must be made is that much of the statistical data in the book is presented in such a way that it is difficult or impossible to decide what conclusions can be drawn. For instance, on page 117 there is a table giving the correlation coefficients of the I.Q.s of pairs of sibs and parents as measured at different times by various authors. No standard errors or sample sizes are shown and one cannot tell whether apparent discrepancies are significant or not. On page 196 there is a discussion of the view that birth injury is more likely at first delivery than in later deliveries. While this view may be correct the evidence given is not adequate to support the theory. Knowledge of the proportion of birth injuries resulting from first pregnancies is not enough : we must also know the proportion of uninjured first births. Again, on page 216, fifteen cases, in which there is parental syphilis as well as juvenile general paresis in the child, are classified in a four-fold table according to the sex of the child and parent. Penrose says " there seems also to be a relative excess of father-son and mother-daughter pairs ". But no statistical appreciation is made. In fact, the exact treatment of the four-fold table gives $P = 4{\cdot}7$ per cent. This is only just significant at 5 per cent. and may well influence the confidence with which we accept the apparent association.

In the foreword Penrose says : " Enough of the historical and *legal* (reviewer's italics) background is explained, I hope, to enable the student properly to appreciate the vicissitudes of the concept of mental deficiency." It is somewhat surprising, after reading this, to find no mention of the M'Naghten ruling and the forensic vicissitudes to which it gives rise, even in the sections on " Psychopathic Personality," " Moral Deficiency " and " Mental Defect and Crime ".

In spite, however, of these criticisms of detail Penrose has here undoubtedly made a valuable contribution to our understanding of the problems of mental defect. There is no short cut, and it is only by a concentrated and sustained attack from many points of view that we shall eventually obtain a solution. As J. B. S. Haldane remarks in his preface, it is greatly to be hoped " that this book will not only be read by physicians, but by social workers and even by one or two of the Members of Parliament and Peers who have to frame the law as to mental defect and criticise its present administration."

NORMAN T. J. BAILEY.

SOVIET GENETICS AND WORLD SCIENCE. By J. S. Huxley. London: Chatto and Windus, 1949. Pp. 245. 8s. 6d.

The genetics controversy between Russia and the West is unique among present-day scientific controversies in its methods as well as in the passionate participation of the lay-public on both sides of the Iron Curtain. These two aspects are closely related. The Russian methods—which consist in an appeal not to scientific argument and facts, but to doctrine and the authority of certain "infallible" authors; in vilification of the moral character of scientific antagonists; and, in the last resort, in persecution—are those in use whenever a new scientific concept challenges cherished traditions and beliefs. In various degrees of severity they have been used against the adherents of Copernicus, against Darwinists and, in Nazi Germany, against Einstein and his disciples.

A special factor of the genetics controversy is that the Russian political orthodoxy tries to give a semblance of scientific authority to their anathema on disliked truths by officially supporting the fantastic theories of Lysenko who, in his ignorance of genetical science and, indeed, of scientific method in general, naïvely believes that he can overthrow the whole edifice of modern genetics by hurling against it a few unproven claims to new discoveries. In fact, it is obvious to every geneticist that those discoveries, even if confirmed by proper scientific experimentation, might at the best mean an interesting extension of our knowledge of heredity, but could in no case shake the well-secured foundation of "Mendel-Morganism."

Indignation against the Russian methods on the one side, loyalty to the Communist dogma on the other, have made it difficult also for Western geneticists to conduct the discussion in that spirit of cold dispassionate objectivity which should prevail in the search for truth; indeed, few if any of them have been capable of so much self-discipline. Having seen the expert rush into the arena without the halo of authority which cold objectivity weaves round his head, the lay public felt entitled to join the issue; in Russia, of course, only on one side, in the West on both. Newspapers and magazines were flooded with letters and articles by persons whose only claim to take part in the discussion often was a passionate political belief. Many of them obviously had not even taken the trouble to read an elementary treatise on genetics; but even scientists of high standing in their own field forgot their customary caution when treading on scientifically unfamiliar ground, and did not hesitate to make pronouncements on genetics which to the expert only revealed lack of knowledge, but to the lay person must have been particularly confusing. The ignorance, the deliberately or unconsciously muddled thinking, the ingenious side-trackings, and the naïve manifestations of wishful thinking revealed in these letters and articles would have made amusing reading, had they not led to the melancholy reflection that the respect of the twentieth century for science and scientific method is only a very thin veneer which all too easily flakes off under emotional stress.

Clearly, a complete, lucid and dispassionate exposition of the whole issue was required. The admirable book by Hudson and Richens is not easily accessible to the general reader and is, in any case, addressed to persons with some genetical knowledge. There can be few who are better suited than Dr Huxley to write a more general account of the subject. He is one of the leading authorities on genetics and evolution; he has

experience in explaining these subjects to students as well as to lay-persons ; he is neither a Communist nor a violent anti-Communist ; he has, in preparation for an article in *Nature*, studied carefully all available sources of information, often comparing different translations of the same Russian text ; he has visited Russia, heard a speech by Lysenko himself, and has had discussions with him as well as with other Russian scientists ; finally, his wide scientific connections have put him in possession of much private information, which in many places lends a pleasantly informal and personal character to the book.

Already in the brief preface, and throughout the book, the hard core of the controversy is deftly disentangled from the manifold and woolly envelopes of side-issues with which it has been surrounded by friend and foe, such as the need of the U.S.S.R. for rapid agricultural progress, or the perversion of Mendelism by the Nazis, or the financial difficulties of Western scientific institutions. To quote Huxley himself : " The major issue at stake (is) not the truth or falsity of Lysenko's claims, but the overriding of science by ideological and political authority." " There is now a party line in genetics. . . . A great scientific nation has repudiated certain basic elements of scientific method, and in so doing has repudiated the universal and supranational character of science. That is the major issue."

The first chapter deals with the nature and history of the controversy and contains a short preliminary characterisation of the two antagonistic conceptions : the scientific disciplines of neo-Mendelism and neo-Darwinism on the one hand, the doctrine of Michurinism on the other. Chapter two is devoted to the ideological issue and is largely taken up by verbatim quotations which abundantly bear out the statements that in the U.S.S.R. " the appeal to fact has been overridden by ideological considerations " and that " scientists (in the U.S.S.R.) who hold certain scientific views can be called names implying that they are unpatriotic or hostile to the political system." This chapter makes depressing reading ; most of all the obsequious recantations of intimidated " converts " from Mendelism to Michurinism, in particular a letter by Zhdanov, printed in full in a postscript. In case the non-scientific reader should be led to the assumption that this state of affairs is usual in heated scientific controversies, Dr Huxley cleverly contrasts each phase of the conflict with what would be expected in a similar controversy among " bourgeois " Western scientists. Chapter three, on " The scientific issue," analyses in detail Lysenko's scientific claims, methods and theories. Chapter four, " Genetics as a science," brings a closely-knit exposition of modern genetics and Neo-Darwinism, with the emphasis on the nature of scientific procedure and argumentation, and states clearly and fairly the reasons why Lamarckism has failed to succeed as a theory of evolution. In the fifth chapter, " The totalitarian regimentation of thought," the Soviet attack on genetics is fitted into the larger picture of the general cultural situation in the U.S.S.R. ; a great number of verbatim quotations show that in many fields other than genetics individual creative or critical activity is subordinated to State and Party doctrine. In this chapter, too, Dr Huxley tries to give an answer to the question, which must have puzzled many scientists among the friends of the Soviet Union, why the Soviet authorities have chosen to support the mystical and ill-founded teachings of Michurin, Prezent and Lysenko to

the acknowledged and progressing science of modern genetics. This is the more astonishing since, as Crane has emphasised, Michurin himself was far removed from the wholesale condemnation of Mendelism which characterises the attitude of his disciples. The last chapter, " The situation of science," discusses briefly how in any modern state an integration of science with other social activities can be reconciled with the autonomy and unity of science. The author's lively interest in education is shown by the fact that a large part of this chapter is taken up with interesting and valuable suggestions for the aims, scope, and methods of a revived form of biological teaching in schools and universities.

The ultimate success of a book on scientific matters depends largely on the answers to two questions: for whom is the book intended? will it prove satisfactory to the class of intended readers? Dr Huxley clearly intends his book for the general educated reader; for he presupposes no knowledge of genetics, biology or scientific procedure in general, and no more knowledge of the Lysenko controversy than any reader of newspapers cannot have helped imbibing. But also the expert, who may think himself thoroughly surfeited with the subject, will find his interest re-awakened by the many new details added in quotations and personal reminiscences. There is, however, the danger that a book which appeals to the expert as well as to the lay-person may, in the end, not satisfy either. In the reviewer's opinion, this danger has not been completely avoided in the present case. In particular it seems that the general reader may be the sufferer from the wide scope of the book. The exposition of genetics and evolution in chapter four may prove somewhat too condensed for him; it also comes too late to help him to an understanding of genetical questions touched upon in the first three chapters. The last two chapters will be of interest to every cultured reader, geneticist or non-geneticist; but from the narrower point of view of the genetics controversy they may weaken the impression made by the first part of the book. There is no doubt that there will be many readers who, for political reasons, are bent on finding excuses for the Russian attitude and on finding fault with Dr Huxley's presentation of it. It will be difficult for them to do so while Huxley is in his own field. By venturing out of it, and especially by bringing in political arguments, Dr Huxley opens the way for just those types of argument which he previously has been at great pains to show up as irrelevant to the major issue. It is, of course, obvious that one of the objects of the book is just this widening of the issue to embrace the whole problem of State interference with any kind of cultural activity: but it would be a great pity if Dr Huxley, by casting his nets too wide, should have reduced the chance of succeeding in his prime purpose which consisted in " dispelling the fog of misunderstanding " which surrounds the genetics controversy.

C. AUERBACH.

DEATH OF A SCIENCE IN RUSSIA. By Conway Zirkle. Philadelphia: University of Pennsylvania Press, 1949. Pp. 319. $3.75.

This book is one of a number of works dealing with the " Genetics Controversy " in the Soviet Union. It contains an exceptionally complete account of the events which have taken place in that country since 1936, culminating in the destruction of the science of genetics in 1948. Gathered

together in one telling volume are the relevant parts of all the documents which bear upon this tragic story. And these documents are accompanied by the thoughtful comments of an intelligent biologist. Those of us who for some years watched the vigorous and spectacular development of genetics in the Soviet Union with high hopes and vicarious pride will be deeply moved by Professor Zirkle's dedication of the book.

"To that great company of Russian geneticists and cytologists, now dispersed and destroyed, to those who lost their positions and are denied the exercise of their profession, to those who simply disappeared, to those who died under mysterious circumstances, to those who, to save their families, recanted : this book is respectfully dedicated."

And so say all of us. S. C. HARLAND.

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